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(71) Applicants *(for all designated States except US)*: **UNIVERSITY OF SHEFFIELD** [GB/GB]; Western Bank, Sheffield S10 2TN (GB). **BIOSYNEXUS INC.** [US/US]; Suite 100, 9610 Medical Centre Drive, Rockville, MD 20850 (US).

(72) Inventors; and

(75) Inventors/Applicants *(for US only)*: **FOSTER, Simon** [GB/GB]; Department of Molecular Biology and Biotechnology, University of Sheffield, Firth Court Western Bank, Sheffield S10 2TN (GB). **McDOWELL, Philip** [GB/GB]; Department of Molecular Biology and Biotechnology, University of Sheffield, Firth Court Western Bank, Sheffield S10 2TN (GB). **BRUMMELL, Kirsty** [GB/GB]; Department of Molecular Biology and Biotechnology, University of Sheffield, Firth Courth Western Bank, Sheffield S10 2TN (GB). **CLARKE, Simon** [GB/GB]; Department of Molecular Biology and Biotechnology, University of Sheffield, Firth Court Western Bank, Sheffield S10 2TN (GB).

(74) Agent: **HARRISON GODDARD FOOTE**; Tower House, Merrion Way, Leeds LS2 8PA (GB).

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(54) Title: ANTIGENIC POLYPEPTIDES

(57) Abstract: The invention relates to a method for the identification of antigenic polypeptides expressed by pathogenic microbes; vaccines comprising said polypeptides; recombinant methods to manufacutre said polypeptides; and therapeutic antibodies directed to said polypeptides.

Antigenic Polypeptides

The invention relates to a method for the identification of antigenic polypeptides expressed by pathogenic microbes; vaccines comprising said polypeptides; recombinant methods to manufacture said polypeptides; and therapeutic antibodies directed to said polypeptides.

Microbial organisms cause a number of fatal or debilitating diseases which affect many millions of people around the world. Currently methods to control microbial organisms include the use of antimicrobial agents (antibiotics) and disinfectants. These have proved to be problematic since exposure to these agents places a significant selection pressure resulting in the creation of resistant microbes which can avoid the effects of the antimicrobial agent(s). For example, recently it has been discovered that microbial organisms have become resistant to triclosan, an agent added to many disinfectants used in households and industrial environments.

An arguably greater problem is the evolution of antibiotic resistant strains of a number of significant pathogenic microbes.

For example, and not by way of limitation, it is estimated that there are up to 50 million people world-wide infected with drug resistant tuberculosis (TB) (Figures from the World Health Organisation, 1998). In the past the use of antibiotics to treat TB relied on the administration of single drugs (eg ethionamide) which promoted a relatively high frequency of resistance. For this reason, combinations of drugs are now used to treat tuberculosis. However the fatality rate in cases caused by strains that are resistant to at least one drug used to treat tuberculosis still approaches 50% even when treatment is given. *Mycobacterium tuberculosis*, the causative agent of TB, is a slow growing bacteria and takes a long time to kill. Therefore, for a drug combination to be effective a person with TB must take the drug combination daily for at least six months. Accordingly, patients frequently have to take two or more pills daily and this requires a regimented dosage over a relatively long period of

treatment. Many patients take the medications only intermittently and therefore do not finish the full course of therapy to completely eradicate the *M. tuberculosis* infection. Moreover, TB is strongly associated with HIV infection and therefore the establishment of TB is strongly correlated with immunosuppression.

Vaccination against TB has been available for many years. The bacillus calmette and guerin (BCG) vaccination has been widely used throughout the world for a long time because it is a safe and inexpensive means to vaccinate large numbers of people who potentially could contract TB. BCG is derived from live, attenuated strains of *Mycobacterium bovis*. However the impact of vaccination on the infectious forms of TB is minimal and BCG has therefore contributed little to the overall control of the disease.

A further example of a pathogenic organism which has developed resistance to antibiotics is *Staphylococcus aureus*. *S.aureus* is a bacterium whose normal habitat is the epithelial lining of the nose in about 20-40% of normal healthy people and is also commonly found on people's skin usually without causing harm. However, in certain circumstances, particularly when skin is damaged, this germ can cause infection. This is a particular problem in hospitals where patients may have surgical procedures and/or be taking immunosuppressive drugs. These patients are much more vulnerable to infection with *S.aureus* because of the treatment they have received. Resistant strains of *S.aureus* have arisen in recent years. Methicillin resistant strains are prevalent and many of these resistant strains are also resistant to several other antibiotics. Currently there is no effective vaccination procedure for *S. aureus*. In the US, *S.aureus* infections are the cause of 13% of the two million hospitalised infections each year. This represents 260,000 people with an infection of *S.aureus*, of which 60-80,000 die.

S. aureus is therefore a major human pathogen capable of causing a wide range of life threatening diseases including septicaemia, endocarditis, arthritis and toxic shock. This ability is determined by the versatility of the organism and its arsenal of

components involved in virulence. Pathogenicity is multifactorial and no one component has shown to be responsible for a particular infection, see Projan, S.J. & Novick, R.P. (1997) in The Staphylococci in Human Disease (Crossley, K.B. & Archer, G.L., eds.) pp.55-81.

At the onset of infection, and as it progresses, the needs and environment of the organism changes and this is mirrored by a corresponding alteration in the virulence determinants which *S. aureus* produces. At the beginning of infection it is important for the pathogen to adhere to host tissues and so a large repertoire of cell surface associated attachment proteins are made. These include collagen-, fibrinogen- and fibronectin-binding proteins. The pathogen also has the ability to evade host defences by the production of factors that reduce phagocytosis or interfere with the ability of the cells to be recognised by circulating antibodies.

Often a focus of infection develops as an abscess and the number of organisms increases. *S. aureus* has the ability to monitor its own cell density by the production of a quorum sensing peptide. Accumulation of the peptide, associated with physiological changes brought about by the beginning of starvation of the cells, elicits a switch in virulence determinant production from adhesins to components involved in invasion and tissue penetration. These include a wide range of hemolysins, proteases and other degradative enzymes.

During the process of any infection the virulence determinants made by *S. aureus* are produced in response to environmental and physiological stimuli. These stimuli will be dependent on the niche within the body and will change as the infection progresses. Little is known of the conditions *in vivo* and it is likely that some components are produced solely in this environment. These are therefore potential vaccine components, which could not be discovered by previous techniques.

One of the most important developments in recent medical history is the development of vaccines which provide prophylactic protection from a wide variety of pathogenic organisms. Many vaccines are produced by inactivated or attenuated pathogens which are injected into an individual. The immunised individual responds by producing both a humoral (antibody) and cellular (cytolytic T cells, CTL's) response. For example, hepatitis vaccines are made by heat inactivating the virus and treating it with a cross linking agent such as formaldehyde. An example of an attenuated pathogen useful as a vaccine is represented by polio vaccines which are produced by attenuating a live pathogen.

However the use of attenuated organisms in vaccines for certain diseases is problematic due to the lack of knowledge regarding the pathology of the condition and the nature of the attenuation. For certain viral agents this is a particular problem since viruses, in particular retroviruses, have an error prone replication cycle which results viable mutations in the genes which comprise the virus. This can result in alterations to antigenic determinants which have previously been used as vaccines. An alternative to the use of inactivated or attenuated pathogens is the identification of pathogen epitopes to which the immune system is particularly sensitive. In this regard many pathogenic toxins produced by pathogenic organisms during an infection are particularly useful in the development of vaccines which protect the individual from a particular pathogenic organism.

The development of so-called subunit vaccines (vaccines in which the immunogen is a fragment or subunit of a protein or complex expressed by a particular pathogenic organism) has been the focus of considerable medical research. The need to identify candidate molecules useful in the development of subunit vaccines is apparent not least because conventional chemotherapeutic approaches to the control of pathogenic organisms has more recently been stymied by the development of antibiotic resistance. A number of methods have been developed to identify potential antigenic polypeptides which can be used as a vaccine. One such method is disclosed herein.

It has been known for many years that tumour cells produce a number of tumour cell specific antigens, some of which are presented at the tumour cell surface. The immune system recognises these antigens as foreign thereby resulting in the production of antibodies to self antigens, so called autoantibodies or autologous antisera.

One such technique is Serological identification of antigens by recombinant Expression Cloning, abbreviated to SEREX.

Typically, the technique involves the extraction of RNA from tumour tissue followed by the selective enrichment of mRNA from the isolated total RNA. The mRNA is reverse transcribed into cDNA using viral reverse transcriptase. The cDNA thus synthesised is subcloned into an expression vector and transformed into an appropriate bacterial strain. The transformed bacteria are plated onto a suitable nutrient agar and under appropriate growth conditions the subcloned cDNA is expressed from the expression vector in the bacterial cell. The cells are lysed naturally by the use of phage based expression vectors, for example λ phage or phagemid based vectors, which through their lytic cycle cause cell lysis. The released polypeptides are transferred to a suitable membrane support (i.e. nitrocellulose, nylon) and exposed to autologous antisera from the patient from which the tumour tissue was originally isolated. The immunoscreening methodology allows the identification of genes that are over expressed or inappropriately expressed in a selected tumour tissue from a patient.

We have exploited this techinque to identify antigenic polypeptides expressed by pathogenic organisms during an infection. Autologous antisera produced during the infection is used to screen an expression library created from genomic DNA to identify and clone antigens.

In its broadest aspect the invention relates to the identification of antigenic polypeptides expressed during an infection by a pathogenic microbe.

According to a first aspect of the invention there is provided a method to identify antigenic polypeptides comprising:

(i) providing a nucleic acid library encoding genes or partial gene sequences of a pathogenic organism;

(ii) transforming/transfecting said library into a host cell;

(iii) providing conditions conducive to the expression of said transformed/transfected genes or partial gene sequences;

(iv) contacting the polypeptides expressed by the genes/partial gene sequences with autologous antisera derived from an animal infected with, or has been infected with, said pathogenic organism; and

(v) purifying the nucleic acid encoding the polypeptide or partial polypeptide binding to said autologous antisera.

In a preferred method of the invention said library comprises genomic DNA of a pathogenic organism.

Ideally said pathogenic organism is bacterial.

More preferably still said bacterial organism is selected from the following: *Staphylococcus aureus; Staphylococcus epidermidis; Enterococcus faecalis; Mycobacterium tuberculsis; Streptococcus group B; Streptoccocus pneumoniae; Helicobacter pylori; Neisseria gonorrhea; Streptococcus group A; Borrelia*

burgdorferi; Coccidiodes immitis; Histoplasma sapsulatum; Neisseria meningitidis type B; Shigella flexneri; Escherichia coli; Haemophilus influenzae.

Preferably still said pathogenic organism is of the genus *Staphylococcus spp.* Ideally organism is *Staphylococcus aureus* or *Staphylococcus epidermidis.*

In a further preferred embodiment of the invention said nucleic acid library is a lambda library, ideally a lambda expression library.

According to a second aspect of the invention there is provided a nucleic acid molecule comprising a DNA sequence selected from:

(i) the DNA sequence as represented in SEQ ID NO's 1 – 13;

(ii) DNA sequences which hybridise to the sequence presented in the SEQ ID No's 1-13 identified in (i) above which encode a polypeptide expressed by a pathogenic organism and

(iii) DNA sequences which are degenerate as a result of the genetic code to the DNA sequences defined in (i) and (ii).

In a yet still further preferred embodiment of the invention said nucleic acid molecule is genomic DNA.

In a preferred embodiment of the invention there is provided an isolated nucleic acid molecule which anneals under stringent hybridisation conditions to the sequences presented in SEQ ID NO's 1- 13.

Stringent hybridisation/washing conditions are well known in the art. For example, nucleic acid hybrids that are stable after washing in 0.1xSSC, 0.1% SDS at 60°C. It

is well known in the art that optimal hybridisation conditions can be calculated if the sequences of the nucleic acid is known. For example, hybridisation conditions can be determined by the GC content of the nucleic acid subject to hybridisation. Please see Sambrook *et al* (1989) Molecular Cloning; A Laboratory Approach. A common formula for calculating the stringency conditions required to achieve hybridisation between nucleic acid molecules of a specified homology is:

$T_m = 81.5^0\,C + 16.6\,Log\,[Na^+] + 0.41[\,\%\,G + C] - 0.63\,(\%formamide)$.

According to a third aspect of the invention there is provided at least one polypeptide identified by the method according to the invention.

In a preferred embodiment of the invention, said polypeptide is associated with infective pathogenicity of an organism according to any previous aspect or embodiment of the invention.

More preferably still said polypeptide is at least one, or part of SEQ ID NO's: 14- 19.

According to a fourth aspect of the invention there is provided a nucleic acid molecule characterised in that said nucleic acid molecule is part of a vector adapted to facilitate recombinant expression of the polypeptide encoded by said nucleic acid molecule.

In a preferred embodiment of the invention said vector is an expression vector adapted for prokaryotic gene expression. Alternatively said expression vector is adapted for eukaryotic gene expression.

Typically said adaptation includes, by example and not by way of limitation, the provision of transcription control sequences (promoter sequences) which mediate cell specific expression. These promoter sequences may be cell specific, inducible or constitutive.

Promoter is an art recognised term and, for the sake of clarity, includes the following features which are provided by example only, and not by way of limitation. Enhancer elements are *cis* acting nucleic acid sequences often found 5' to the transcription initiation site of a gene (enhancers can also be found 3' to a gene sequence or even located in intronic sequences and is therefore position independent). Enhancers function to increase the rate of transcription of the gene to which the enhancer is linked. Enhancer activity is responsive to *trans* acting transcription factors (polypeptides) which have been shown to bind specifically to enhancer elements. The binding/activity of transcription factors (please see Eukaryotic Transcription Factors, by David S Latchman, Academic Press Ltd, San Diego) is responsive to a number of environmental cues which include, by example and not by way of limitation, intermediary metabolites (eg glucose, lipids), environmental effectors (eg light, heat,).

Promoter elements also include so called TATA box and RNA polymerase initiation selection (RIS) sequences which function to select a site of transcription initiation. These sequences also bind polypeptides which function, *inter alia*, to facilitate transcription initiation selection by RNA polymerase.

Adaptations also include the provision of selectable markers and autonomous replication sequences which both facilitate the maintenance of said vector in either the eukaryotic cell or prokaryotic host. Vectors which are maintained autonomously are referred to as episomal vectors.

Adaptations which facilitate the expression of vector encoded genes include the provision of transcription termination/polyadenylation sequences. This also includes the provision of internal ribosome entry sites (IRES) which function to maximise expression of vector encoded genes arranged in bicistronic or multi-cistronic expression cassettes.

These adaptations are well known in the art. There is a significant amount of published literature with respect to expression vector construction and recombinant DNA techniques in general. Please see, Sambrook et al (1989) Molecular Cloning: A Laboratory Manual, Cold Spring Harbour Laboratory, Cold Spring Harbour, NY and references therein; Marston, F (1987) DNA Cloning Techniques: A Practical Approach Vol III IRL Press, Oxford UK; DNA Cloning: F M Ausubel et al, Current Protocols in Molecular Biology, John Wiley & Sons, Inc.(1994).

According to yet a further aspect of the invention there is provided a method for the production of the polypeptides according to any previous aspect or embodiment of the invention comprising:

(i) providing a cell transformed/transfected with a vector according to the invention;

(ii) growing said cell in conditions conducive to the manufacture of said polypeptides; and

(iii) purifying said polypeptide from said cell, or its growth environment.

In a preferred method of the invention said vector encodes, and thus said recombinant polypeptide is provided with, a secretion signal to facilitate purification of said polypeptide.

According to a fifth aspect of the invention there is provided a cell or cell-line transformed or transfected with the vector according to the invention.

In a preferred embodiment of the invention said cell is a prokaryotic cell. Alternatively said cell is a eukaryotic cell selected from: fungal, insect, amphibian; mammalian; plant.

According to a yet further aspect of the invention there is provided a vaccine comprising at least one polypeptide according to the invention.

Ideally said vaccine further comprises a carrier and/or adjuvant.

The terms adjuvant and carrier are construed in the following manner. Some polypeptide or peptide antigens contain B-cell epitopes but no T cell epitopes. Immune responses can be greatly enhanced by the inclusion of a T cell epitope in the polypeptide/peptide or by the conjugation of the polypeptide/peptide to an immunogenic carrier protein such as key hole limpet haemocyanin or tetanus toxoid which contain multiple T cell epitopes. The conjugate is taken up by antigen presenting cells, processed and presented by human leukocyte antigens (HLA's) class II molecules. This allows T cell help to be given by T cell's specific for carrier derived epitopes to the B cell which is specific for the original antigenic polypeptide/peptide. This can lead to increase in antibody production, secretion and isotype switching.

An adjuvant is a substance or procedure which augments specific immune responses to antigens by modulating the activity of immune cells. Examples of adjuvants include, by example only, agonsitic antibodies to co-stimulatory molecules, Freunds adjuvant, muramyl dipeptides, liposomes. An adjuvant is therefore an immunomodulator. A carrier is an immunogenic molecule which, when bound to a second molecule augments immune responses to the latter.

In yet a further aspect of the invention there is provided a method to immunise an animal against a pathogenic microbe comprising administering to said animal at least one polypeptide, or part thereof, according to the invention or the vaccine according to the invention.

In a preferred method of the invention said animal is human.

Preferably the vaccine, or antigenic polypeptide, can be delivered by direct injection either intravenously, intramuscularly, subcutaneously. Further still, the vaccine or antigenic polypeptide, may be taken orally.

Preferably the vaccine is against the bacterial species *Staphylococcus aureus.*

The vaccine may also be against the bacterial species *Staphylococcus epidermidis.*

It will also be apparent that vaccines or antigenic polypeptides are effective at preventing or alleviating conditions in animals other than humans, for example and not by way of limitation, family pets, livestock, horses.

According to a further aspect of the invention there is provided an antibody, or at least an effective binding part thereof, which binds at least one polypeptide according to the invention.

In a preferred embodiment of the invention said antibody is a polyclonal or monoclonal antibody wherein said antibody is specific to said polypeptide.

Alternatively, said antibody is a chimeric antibody produced by recombinant methods to contain the variable region of said antibody with an invariant or constant region of a human antibody.

In a further alternative embodiment of the invention, said antibody is humanised by recombinant methods to combine the complimentarity determining regions of said antibody with both the constant (C) regions and the framework regions from the variable (V) regions of a human antibody.

Preferably said antibody is provided with a marker including a conventional label or tag, for example a radioactive and/or fluorescent and/or epitope label or tag.

Preferably said humanised monoclonal antibody to said polypeptide is produced as a fusion polypeptide in an expression vector suitably adapted for transfection or transformation of prokaryotic or eukaryotic cells.

Antibodies, also known as immunoglobulins, are protein molecules which have specificity for foreign molecules (antigens). Immunoglobulins (Ig) are a class of structurally related proteins consisting of two pairs of polypeptide chains, one pair of light (L) (low molecular weight) chain (κ or λ), and one pair of heavy (H) chains (γ, α, μ, δ and ε), all four linked together by disulphide bonds. Both H and L chains have regions that contribute to the binding of antigen and that are highly variable from one Ig molecule to another. In addition, H and L chains contain regions that are non-variable or constant.

The L chains consist of two domains. The carboxy-terminal domain is essentially identical among L chains of a given type and is referred to as the "constant" (C) region. The amino terminal domain varies from L chain to L chain and contributes to the binding site of the antibody. Because of its variability, it is referred to as the "variable" (V) region.

The H chains of Ig molecules are of several classes, α, μ, σ, α, and γ (of which there are several sub-classes). An assembled Ig molecule consisting of one or more units of two identical H and L chains, derives its name from the H chain that it possesses. Thus, there are five Ig isotypes: IgA, IgM, IgD, IgE and IgG (with four sub-classes based on the differences in the H chains, i.e., IgG1, IgG2, IgG3 and IgG4). Further detail regarding antibody structure and their various functions can be found in, Using Antibodies: A laboratory manual, Cold Spring Harbour Laboratory Press.

Chimeric antibodies are recombinant antibodies in which all of the V-regions of a mouse or rat antibody are combined with human antibody C-regions. Humanised antibodies are recombinant hybrid antibodies which fuse the complimentarity determining regions from a rodent antibody V-region with the framework regions from the human antibody V-regions. The C-regions from the human antibody are also used. The complimentarity determining regions (CDRs) are the regions within the N-terminal domain of both the heavy and light chain of the antibody to where the

majority of the variation of the V-region is restricted. These regions form loops at the surface of the antibody molecule. These loops provide the binding surface between the antibody and antigen.

Antibodies from non-human animals provoke an immune response to the foreign antibody and its removal from the circulation. Both chimeric and humanised antibodies have reduced antigenicity when injected to a human subject because there is a reduced amount of rodent (i.e. foreign) antibody within the recombinant hybrid antibody, while the human antibody regions do not illicit an immune response. This results in a weaker immune response and a decrease in the clearance of the antibody. This is clearly desirable when using therapeutic antibodies in the treatment of human diseases. Humanised antibodies are designed to have less "foreign" antibody regions and are therefore thought to be less immunogenic than chimeric antibodies.

In another aspect of the invention there is provided a vector which is adapted for the expression of the humanised or chimeric antibodies according to the invention.

In a yet further aspect of the invention, there is provided a cell or cell line which has been transformed or transfected with the vector encoding the humanised or chimeric antibody according to the invention.

In a yet further aspect of the invention there is provided a method for the production of the humanised or chimeric antibody according to the invention comprising :

(i) providing a cell transformed or transfected with a vector which comprises a nucleic acid molecule encoding the humanised or chimeric antibody according to the invention;

(ii) growing said cell in conditions conducive to the manufacture of said antibody; and

(iii) purifying said antibody from said cell, or its growth environment.

In a yet further aspect of the invention there is provided a hybridoma cell line which produces a monoclonal antibody as hereinbefore described.

In a further aspect of the invention there is provided a method of producing monoclonal antibodies according to the invention using hybridoma cell lines according to the invention.

In a further aspect of the invention there is provided a method for preparing a hybridoma cell-line producing monoclonal antibodies according to the invention comprising the steps of:

i) immunising an immunocompetent mammal with an immunogen comprising at least one polypeptide having the amino acid sequence as represented in SEQ. ID No 14-19, or fragments thereof;

ii) fusing lymphocytes of the immunised immunocompetent mammal with myeloma cells to form hybridoma cells;

iii) screening monoclonal antibodies produced by the hybridoma cells of step (ii) for binding activity to the amino acid sequences of (i);

iv) culturing the hybridoma cells to proliferate and/or to secrete said monoclonal antibody; and

v) recovering the monoclonal antibody from the culture supernatant.

Preferably, the said immunocompetent mammal is a mouse. Alternatively, said immunocompetent mammal is a rat.

The production of monoclonal antibodies using hybridoma cells is well-known in the art. The methods used to produce monoclonal antibodies are disclosed by Kohler and Milstein in Nature 256, 495-497 (1975) and also by Donillard and Hoffman, "Basic Facts about Hybridomas" in Compendium of Immunology V.II ed. by Schwartz, 1981, which are incorporated by reference.

In a further aspect of the invention there is provided the use of the antibodies for manufacture of a medicament for the treatment of *Staphylococcus aureus*-associated septicaemia, food-poisoning or skin disorders.

In another aspect of the invention there is provided the use of the antibodies according to the invention for the manufacture of a medicament for the treatment of *Staphylococcus epidermidis*-associated septicaemia, peritonitis or endocarditis.

It will be apparent that the polypeptides identified by the method according to the invention will facilitate the production of therapeutic antibodies to a range of diseases resulting from pathogenic infection, for example, septicaemia; tuberculosis; bacteria-associated food poisoning; blood infections; peritonitis; endocarditis; sepsis; meningitis; pneumonia; stomach ulcers; gonorrhoea; strep throat; streptococcal-associated toxic shock; necrotizing fasciitis; impetigo; histoplasmosis; Lyme disease; gastro-enteritis; dysentery; shigellosis.

As has already been stated earlier, microbial organisms cause a wide variety of diseases. Listed below, and not by way of limitation, are a number of micro-organisms and some of the diseases they cause.

Micro-organism	**Disease(s) caused**
Staphylococcus aureus	Sepsis, food poisoning, septicaemia,
Staphylococcus epidermidis	Peritonitis, septicaemia, endocarditis, other hospital-associated diseases
Enterococcus faecalis	Endocarditis, cystitis, wound infections
Mycobacterium tuberculosis	Tuberculosis
Streptococcus group B	Sepsis, meningitis, pneumonia, bladder infections
Streptococcus pneumoniae	Pneumonia, meningitis
Helicobacter pylori	Stomach ulcers
Neisseria gonorrhoea	Gonorrhoea
Streptococcus group A	Strep throat, necrotizing fasciitis, impetigo, Strep. Toxic shock syndrome
Borrelia burgdoferi	Lyme disease
Coccidiodes immitis	Pneumonia

Histoplasma sapsulatum	Histoplasmosis, pneumonia
Neisseria meningitidis type B	Meningitis
Shigella flexneri	Gastro-enteritis, shigellosis, dysentry
Escherichia coli	Food-poisoning, gastro-enteritis
Haemophilus influenzae	Meningitis, pneumonia, arthritis, cellulitis

An embodiment of the invention will now be described by example only and with reference to the following materials, methods and SEQ ID NO's 1 –19 and Table 1.

Materials and Methods

A λZAP Express library of genomic DNA of *S. aureus* 8325/4 was used. It contains fragments of 2-10kb from a partial *Sau*3A digest of total genomic DNA. This was cloned into the *Bam*H1 site of the vector. The library contains >10x coverage of the genome. The library was probed by plaque lift using an initial screen of approximately 20,000 plaque forming units on a 9cm diameter Petri dish. The plating cells used, their treatment, the plating procedure and buffers were exactly as described in the manufacturers handbook (Stratagene). Plating cells, *Escherichia coli* XL1-Blue MRF', were infected with phage and plated in 3 ml top LB agar containing 10 mM $MgSO_4$ onto LB plates containing 10 mM $MgSO_4$. The plates were then incubated at 42°C for 4 hr. An 8.5cm diameter nitrocellulose filter disc (previously soaked in 10 mM IPTG and air-dried) was placed on each plate and its location marked. The plates were then incubated for a further 3.5 hr at 37°C. The filters were removed and washed in TBST buffer before blocking overnight at 4°C in TBST containing 6% w/v dried skimmed milk and 3% v/v pig serum (Sigma). The serum was used to block any Protein A clones on the filter. The filters are then treated with patient serum (1/5000 dilution) in blocking solution for 90 min at room temperature. Antisera have been obtained from patients convalescing from major *S. aureus* infections. The filters are then washed for 3x10 min in TBST. Secondary antibody used was goat anti-human whole IgG alkaline phosphatase linked (Sigma)

at 1/30,000 dilution in blocking solution at room temperature for 30 min. The filters were then washed as above and developed using a standard colorimetric procedure.

Cross-reactive plaques were located on the agar plates and cored into 0.2ml phage buffer with 0.02 ml chloroform. The titre of each core stock was determined and the phage plated at approximately 200 plaques per plate. A plaque lift and screen was performed as above to give single, pure cross-reactive clones.

The pure clones were then spotted (1μl) onto plates to give a confluent plaque of 0.5cm diameter. 30 individual clones can be spotted on each plate. A plaque lift is performed and the filter probed with an appropriate sera. In this way clones can be tested for their cross-reactivity with other patient sera, non-infected donor sera and anti-Protein A sera.

Individual clones were then excised to give a phagemid in *E. coli* XLOLR using the manufacturers protocol (Stratagene). A plasmid miniprep of each was carried out and the size of the genomic insert determined by restriction mapping. The identity of the cloned insert was determined by DNA sequencing using primers against vector sequence, which allows sequencing across the insert. By comparison of the derived sequence against the public domain databases the nature of the cloned gene(s) can be determined.

Hybridisation Solutions/Conditions

Typically, hybridisation conditions uses 4 – 6 x SSPE (20x SSPE contains 175.3g NaCl, 88.2g NaH_2PO_4 H_2O and 7.4g EDTA dissolved to 1 litre and the pH adjusted to 7.4); 5-10x Denhardts solution (50x Denhardts solution contains 5g Ficoll (type 400, Pharmacia), 5g polyvinylpyrrolidone abd 5g bovine serum albumen; 100μg-1.0mg/ml sonicated salmon/herring DNA; 0.1-1.0% sodium dodecyl sulphate; optionally 40-60% deionised formamide. Hybridisation temperature will vary

depending on the GC content of the nucleic acid target sequence but will typically be between 42^0- 65^0 C.

Staphylococcus aureus clones identified in human sera screen

TABLE 1

Patient Sera	Clone	Encoded proteins	Locus number
A	1	γ hemolysin B and C subunit	1
A	3	Atl	2
A	4	γ hemolysin B and C subunit	1
A	5	γ hemolysin B and C subunit	1
A	7	Novel putative protease (ORF1 novel antigen like)	7
A	8	Novel nuclease (YisK)	5
A	9	Novel autolysin	6
A	10	γ hemolysin B and C subunit	1
A	11	Atl	2
A	14	γ hemolysin B and C subunit	1
A	15	γ hemolysin B and C subunit	1
A	S1	Novel putative protease (ORF1 novel antigen like)	7
A	S5	Novel surface protein	12
A	S17	γ hemolysin B and C subunit	1
A	S18	Novel putative protease (ORF1 novel antigen like)	7
A	S19	Novel autolysin	6
A	S20	Novel surface protein/toxin	13
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F	1	Novel exotoxin (exotoxin 2 like)	8
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F	3	Novel exotoxin (exotoxin 2 like)	8
F	4	Novel exotoxin (exotoxin 2 like)	8
F	5	Novel hemolysin (YjfD)	11

CLAIMS

1. An isolated nucleic acid molecule comprising a DNA sequence selected from the group consisting of:

(i) the DNA sequence as represented in SEQ ID NO's 1 – 13;

(ii) DNA sequences which hybridise to the sequence presented in the SEQ ID No's 1-13 identified in (i) above and which encode a polypeptide expressed by a pathogenic organism; and

(iii) DNA sequences which are degenerate as a result of the genetic code to the DNA sequences defined in (i) and (ii).

2. An isolated nucleic acid molecule according to claim 1 which is genomic DNA.

3. An isolated nucleic acid molecule according to claim 1 or 2 which anneals under stringent hybridisation conditions to the sequences presented in SEQ ID NO's 1-13.

4. A vector comprising a nucleic acid molecule according to any of claims 1-3.

5. A vector according to claim 4 wherein the vector is adapted for recombinant expression of the polypeptide encoded by the nucleic acid.

6. A vector according to claim 4 or 5 wherein said vector is an expression vector adapted for prokaryotic gene expression.

7. A vector according to claim 4 or 5 wherein said vector is an expression vector adapted for eukaryotic gene expression.

8. A vector according to any of claims 4 to 7 wherein the adaptation of the vector includes the provision of promoter sequences.

9. A vector according to claim 8 wherein the promoter sequences provide for cell specific, inducible or constitutive expression.

10. A method to identify antigenic polypeptides comprising:

(i) providing a nucleic acid library encoding genes or partial gene sequences of a pathogenic organism;

(ii) transforming/transfecting said library into a host cell;

(iii) contacting the polypeptides expressed by the genes/partial gene sequences with autologous antisera derived from an animal infected with, or has been infected with, said pathogenic organism; and

(iv) purifying the nucleic acid encoding the polypeptide or partial polypeptide binding to said autologous antisera.

11. A method according to claim 10 wherein said library comprises genomic DNA of a pathogenic organism.

12. A method according to claim 10 or claim 11 wherein said pathogenic organism is bacterial.

13. A method according to any of claims 10 to 12 wherein said bacterial organism is selected from the following: *Staphylococcus aureus; Staphylococcus epidermidis; Enterococcus faecalis; Mycobacterium tuberculsis; Streptococcus group B; Streptoccocus pneumoniae; Helicobacter pylori;*

Neisseria gonorrhea; Streptococcus group A; Borrelia burgdorferi; Coccidiodes immitis; Histoplasma sapsulatum; Neisseria meningitidis type B; Shigella flexneri; Escherichia coli; Haemophilus influenzae

14. A method according to any of claim 13 wherein said pathogenic organism is *Staphylococcus aureus*.

15. A method according to any of claim 13 wherein said pathogenic organism is *Staphylococcus epidermidis*.

16. A method according to any of claims 10 to 15 wherein said nucleic acid library is a lambda library.

17. A polypeptide identified by the method according to any of claims 10 to 16.

18. A polypeptide according to claim 17 which is selected from the group consisting of SEQ ID NO's: 14-19.

19. A method for the production of the polypeptides according to any of claims 17 or 18 comprising:

(i) providing a cell transformed/transfected with a vector according to any of claims 4 to 9 and with cell culture conditions; and

(ii) purifying said polypeptide from said cell, or its growth environment.

20. A method according to claim 19 wherein said vector encodes, and thus said recombinant polypeptide is provided with, a secretion signal to facilitate purification of said polypeptide.

21. A cell transformed or transfected with the vector according to any of claims 4 to 9.

22. A cell according to claim 21 which is a prokaryotic cell.

23. A cell according to claim 21 which is a eukaryotic cell selected from the group consisting of: fungal cell, insect cell, amphibian cell; mammalian cell; plant cell.

24. A vaccine comprising at least one polypeptide according to claims 16 or 17.

25. A vaccine according to claim 24 which further comprises a carrier and/or adjuvant.

26. A method to immunise an animal against a pathogenic microbe comprising administering to the animal at least one polypeptide, or part thereof, according to any previous claim or the vaccine of any previous claim.

27. A method according to claim 26 wherein the animal is human.

28. A method according to claim 26 or 27 wherein the vaccine, or antigenic polypeptide, is delivered by direct injection either intravenously, intramuscularly or subcutaneously.

29. A method according to claim 25 or 26 wherein the vaccine or antigenic polypeptide is taken orally.

30. A method according to any of claims 26 to 29 wherein the vaccine is against the bacterial genus *Staphylococcus spp.*

31. A method according to claim 30 wherein the vaccine is against the bacterial species *Staphylococcus aureus.*

32. A method according to claim 30 wherein the vaccine is against the bacterial species *Staphylococcus epidermidis.*

33. An antibody, or at least an effective part thereof, which binds at least with a selective part of the polypeptide according to claim 16 or 17.

34. An antibody according to claim 33 which is a monoclonal antibody.

35. An antibody according to claim 33 or 34 wherein said effective part comprises FAb fragments.

36. An antibody according to any of claims 33 to 35 which is a chimeric antibody.

37. An antibody according to any of claims 33 to 35 which is a humanised antibody.

38. An antibody according to any of claims 33 to 37 wherein said antibody is provided with a marker, label or tag.

39. An antibody according to claim 38 wherein said antibody is provided with a marker selected from a group consisting of: a radioactive label, a fluorescent label; an epitope tag.

40. An antibody according to any of claims 34 to 39 which is produced as a fusion polypeptide.

41. A vector which is adapted for the expression of the antibodies according to any of claims 34-40.

42. A cell which has been transformed or transfected with the vector according to claim 41.

43. A method for the production of the antibody according to any of claims 34 or 40 comprising :

i) providing a cell transformed or transfected with the vector according to claim 41 and with cell culture conditions; and

ii) purifying said antibody from said cell, or its growth environment.

44. A hybridoma cell line which produces an antibody according to claim 34.

45. Use of the antibodies according to any of claims 33 to 40 for the manufacture of a medicament for the treatment of *Staphylococcus aureus*-associated septicaemia, food-poisoning or skin disorders.

46. Use of the antibodies according to any of claims 33 to 40 for the manufacture of a medicament for the treatment of *Staphylococcus epidermidis*-associated septicaemia, peritonitis or endocarditis

47. A method for preparing a hybridoma cell-line producing monoclonal antibodies according to claim 34, comprising the steps of:

i) immunising an immunocompetent mammal with an immunogen comprising at least one polypeptide having the amino acid sequence as set forward in SEQ ID No: 14-19, or fragments thereof;

ii) fusing lymphocytes of the immunised immunocompetent mammal with myeloma cells to form hybridoma cells;

iii) screening monoclonal antibodies produced by the hybridoma cells of step (ii) for binding activity to the amino acid sequences of (i);

iv) culturing the hybridoma cells to proliferate and/or to secrete said monoclonal antibody; and

v) recovering the monoclonal antibody from the culture supernatant.

48. A method according to claim 47, wherein said immunocompetent mammal is a mouse

49. A method according to claim 47, wherein said immunocompetent mammal is a rat

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<211> 2794
<212> DNA
<213> Staphylococcus aureus

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<211> 673
<212> DNA
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Lys Gln Val Ser Val Gly Lys Asp Val Tyr Leu Tyr Gly Thr Ile Asn
610 615 620

Asn Arg Thr Gly Trp Val Asn Ala Lys Asp Leu Thr Ala Pro Thr Ala
625 630 635 640

Val Lys Pro Thr Thr Ser Ala Ala Lys Asp Tyr Asn Tyr Thr Tyr Val

645 650 655

Ile Lys Asn Gly Asn Gly Tyr Tyr Tyr Val Thr Pro Asn Ser Asp Thr
660 665 670

Ala Lys Tyr Ser Leu Lys Ala Phe Asn Glu Gln Pro Phe Ala Val Val
675 680 685

Lys Glu Gln Val Ile Asn Gly Gln Thr Trp Tyr Tyr Gly Lys Leu Ser
690 695 700

Asn Gly Lys Leu Ala Trp Ile Lys Ser Thr Asp Leu Ala Lys Glu Leu
705 710 715 720

Ile Lys Tyr Asn Gln Thr Gly Met Ala Leu Asn Gln Val Ala Gln Ile
725 730 735

Gln Ala Gly Leu Gln Tyr Lys Pro Gln Val Gln Arg Val Pro Gly Lys
740 745 750

Trp Thr Gly Ala Asn Phe Asn Asp Val Lys His Ala Met Asp Thr Lys
755 760 765

Arg Leu Ala Gln Asp Pro Ala Leu Lys Tyr Gln Phe Leu Arg Leu Asp
770 775 780

Gln Pro Gln Asn Ile Ser Ile Asp Lys Ile Asn Gln Phe Leu Lys Gly
785 790 795 800

Lys Gly Val Leu Glu Asn Gln Gly Ala Ala Phe Asn Lys Ala Ala Gln
805 810 815

Met Tyr Gly Ile Asn Glu Val Tyr Leu Ile Ser His Ala Leu Leu Glu
820 825 830

Thr Gly Asn Gly Thr Ser Gln Leu Ala Lys Gly Ala Asp Val Val Asn
835 840 845

Asn Lys Val Val Thr Asn Ser Asn Thr Lys Tyr His Asn Val Phe Gly
850 855 860

Ile Ala Ala Tyr Asp Asn Asp Pro Leu Arg Glu Gly Ile Lys Tyr Ala
865 870 875 880

Lys Gln Ala Gly Trp Asp Thr Val Ser Lys Ala Ile Val Gly Gly Ala
885 890 895

Lys Phe Ile Gly Asn Ser Tyr Val Lys Ala Gly Gln Asn Thr Leu Tyr
900 905 910

Lys Met Arg Trp Asn Pro Ala His Pro Gly Thr His Gln Tyr Ala Thr
915 920 925

Asp Val Asp Trp Ala Asn Ile Asn Ala Lys Ile Ile Lys Gly Tyr Tyr
930 935 940

Asp Lys Ile Gly Glu Val Gly Lys Tyr Phe Asp Ile Pro Gln Tyr Lys
945 950 955 960

<210> 16
<211> 386
<212> PRT
<213> Staphylococcus aureus

<400> 16

```
Asp Gln Tyr Ser Glu Asp Ala Lys Lys Thr Gln Lys Asp Tyr Ala Ser
  1               5                  10                  15

Gln Ser Lys Lys Asp Lys Asn Glu Lys Ser Asn Thr Lys Asn Pro Gln
             20                  25                  30

Leu Pro Thr Gln Asp Glu Leu Lys His Lys Ser Lys Pro Ala Gln Ser
         35                  40                  45

Phe Asn Asn Asp Val Asn Gln Lys Asp Thr Arg Ala Thr Ser Leu Phe
     50                  55                  60

Glu Thr Asp Pro Ser Ile Ser Asn Asn Asp Asp Ser Gly Gln Phe Asn
 65                  70                  75                  80

Val Val Asp Ser Lys Asp Thr Arg Gln Phe Val Lys Ser Ile Ala Lys
                 85                  90                  95

Asp Ala His Arg Ile Gly Gln Asp Asn Asp Ile Tyr Ala Ser Val Met
            100                 105                 110

Ile Ala Gln Ala Ile Leu Glu Ser Asp Ser Gly Arg Ser Ala Leu Ala
        115                 120                 125

Lys Ser Pro Asn His Asn Leu Phe Gly Ile Lys Gly Ala Phe Glu Gly
    130                 135                 140

Asn Ser Val Pro Phe Asn Thr Leu Glu Ala Asp Gly Asn Gln Leu Tyr
145                 150                 155                 160

Ser Ile Asn Ala Gly Phe Arg Lys Tyr Pro Ser Thr Lys Glu Ser Leu
                165                 170                 175

Lys Asp Tyr Ser Asp Leu Ile Lys Asn Gly Ile Asp Gly Asn Arg Thr
            180                 185                 190

Ile Tyr Lys Pro Thr Trp Lys Ser Glu Ala Asp Ser Tyr Lys Asp Ala
        195                 200                 205

Thr Ser His Leu Ser Lys Thr Tyr Ala Thr Asp Pro Asn Tyr Ala Lys
    210                 215                 220

Lys Leu Asn Ser Ile Ile Lys His Tyr Gln Leu Thr Gln Phe Asp Asp
225                 230                 235                 240

Glu Arg Met Pro Asp Leu Asp Lys Tyr Glu Arg Ser Ile Lys Asp Tyr
                245                 250                 255

Asp Asp Ser Ser Asp Glu Phe Lys Pro Phe Arg Glu Val Ser Asp Ser
            260                 265                 270

Met Pro Tyr Pro His Gly Gln Cys Thr Trp Tyr Val Tyr Asn Arg Met
        275                 280                 285

Lys Gln Phe Gly Thr Ser Ile Ser Gly Asp Leu Gly Asp Ala His Asn
    290                 295                 300
```

```
Trp Asn Asn Arg Ala Gln Tyr Arg Asp Tyr Gln Val Ser His Thr Pro
305                 310                 315                 320

Lys Arg His Ala Ala Val Val Phe Glu Ala Gly Gln Phe Gly Ala Asp
                325                 330                 335

Gln His Tyr Gly His Val Ala Phe Val Glu Lys Val Asn Ser Asp Gly
            340                 345                 350

Ser Ile Val Ile Ser Glu Ser Asn Val Lys Gly Leu Gly Ile Ile Ser
        355                 360                 365

His Arg Thr Ile Asn Ala Ala Ala Ala Glu Glu Leu Ser Tyr Ile Thr
    370                 375                 380

Gly Lys
385
```

<210> 17
<211> 325
<212> PRT
<213> Staphylococcus aureus

<400> 17

```
Met Lys Met Asn Lys Leu Val Lys Ser Ser Val Ala Thr Ser Met Ala
  1               5                  10                  15

Leu Leu Leu Leu Ser Gly Thr Ala Asn Ala Glu Gly Lys Ile Thr Pro
             20                  25                  30

Val Ser Val Lys Lys Val Asp Asp Lys Val Thr Leu Tyr Lys Thr Thr
         35                  40                  45

Ala Thr Ala Asp Ser Asp Lys Phe Lys Ile Ser Gln Ile Leu Thr Phe
     50                  55                  60

Asn Phe Ile Lys Asp Lys Ser Tyr Asp Lys Asp Thr Leu Val Leu Lys
 65                  70                  75                  80

Ala Thr Gly Asn Ile Asn Ser Gly Phe Val Lys Pro Asn Pro Asn Asp
                 85                  90                  95

Tyr Asp Phe Ser Lys Leu Tyr Trp Gly Ala Lys Tyr Asn Val Ser Ile
            100                 105                 110

Ser Ser Gln Ser Asn Asp Ser Val Asn Val Val Asp Tyr Ala Pro Lys
        115                 120                 125

Asn Gln Asn Glu Glu Phe Gln Val Gln Asn Thr Leu Gly Tyr Thr Phe
    130                 135                 140

Gly Gly Asp Ile Ser Ile Ser Asn Gly Leu Ser Gly Gly Leu Asn Gly
145                 150                 155                 160

Asn Thr Ala Phe Ser Glu Thr Ile Asn Tyr Lys Gln Glu Ser Tyr Arg
                165                 170                 175

Thr Thr Leu Ser Arg Asn Thr Asn Tyr Lys Asn Val Gly Trp Gly Val
            180                 185                 190

Glu Ala His Lys Ile Met Asn Asn Gly Trp Gly Pro Tyr Gly Arg Asp
        195                 200                 205
```

```
Ser Phe His Pro Thr Tyr Gly Asn Glu Leu Phe Leu Ala Gly Arg Gln
    210                 215                 220

Ser Ser Ala Tyr Ala Gly Gln Asn Phe Ile Ala Gln His Gln Met Pro
225                 230                 235                 240

Leu Leu Ser Arg Ser Asn Phe Asn Pro Glu Phe Leu Ser Val Leu Ser
                245                 250                 255

His Arg Gln Asp Gly Ala Lys Lys Ser Lys Ile Thr Val Thr Tyr Gln
            260                 265                 270

Arg Glu Met Asp Leu Tyr Gln Ile Arg Trp Asn Gly Phe Tyr Trp Ala
        275                 280                 285

Gly Ala Asn Tyr Lys Asn Phe Lys Thr Arg Thr Phe Lys Ser Thr Tyr
    290                 295                 300

Glu Ile Asp Trp Glu Asn His Lys Val Lys Leu Leu Asp Thr Lys Glu
305                 310                 315                 320

Thr Glu Asn Asn Lys
                325
```

<210> 18
<211> 157
<212> PRT
<213> Staphylococcus aureus

<400> 18

```
Ser Phe Asn Tyr Ser Lys Ser Ile Ser Tyr Thr Gln Gln Asn Tyr Val
  1               5                  10                  15

Ser Glu Val Glu Gln Gln Asn Ser Lys Ser Val Leu Trp Gly Val Lys
            20                  25                  30

Ala Asn Ser Phe Ala Thr Glu Ser Gly Gln Lys Ser Ala Phe Asp Ser
        35                  40                  45

Asp Leu Phe Val Gly Tyr Lys Pro His Ser Lys Asp Pro Arg Asp Tyr
    50                  55                  60

Phe Val Pro Asp Ser Glu Leu Pro Pro Leu Val Gln Ser Gly Phe Asn
65                  70                  75                  80

Pro Ser Phe Ile Ala Thr Val Ser His Glu Lys Gly Ser Ser Asp Thr
                85                  90                  95

Ser Glu Phe Glu Ile Thr Tyr Gly Arg Asn Met Asp Val Thr His Ala
            100                 105                 110

Ile Lys Arg Ser Thr His Tyr Gly Asn Ser Tyr Leu Asp Gly His Arg
        115                 120                 125

Val His Asn Ala Phe Val Asn Arg Asn Tyr Thr Val Lys Tyr Glu Val
    130                 135                 140

Asn Trp Lys Thr His Glu Ile Lys Val Lys Gly Gln Asn
145                 150                 155
```

<210> 19
<211> 345
<212> PRT
<213> Staphylococcus aureus

<400> 19

```
Ile Ile Ala Ile Ile Ile Leu Ile Phe Ile Ser Phe Phe Phe Ser Gly
  1               5                  10                  15

Ser Glu Thr Ala Leu Thr Ala Ala Asn Lys Ala Lys Phe Lys Thr Glu
             20                  25                  30

Ala Asp Lys Gly Asp Lys Lys Ala Lys Gly Ile Val Lys Leu Leu Glu
         35                  40                  45

Lys Pro Ser Glu Phe Ile Thr Thr Ile Leu Ile Gly Asn Asn Val Ala
     50                  55                  60

Asn Ile Leu Leu Pro Thr Leu Val Thr Ile Met Ala Leu Arg Trp Gly
 65                  70                  75                  80

Ile Ser Val Gly Ile Ala Ser Ala Val Leu Thr Val Val Ile Ile Leu
                 85                  90                  95

Ile Ser Glu Val Ile Pro Lys Ser Val Ala Ala Thr Phe Pro Asp Lys
            100                 105                 110

Ile Thr Arg Leu Val Tyr Pro Ile Ile Asn Ile Cys Val Ile Val Phe
        115                 120                 125

Arg Pro Ile Thr Leu Leu Leu Asn Lys Leu Thr Asp Ser Ile Asn Arg
    130                 135                 140

Ser Leu Ser Lys Gly Gln Pro Gln Glu His Gln Phe Ser Lys Glu Glu
145                 150                 155                 160

Phe Lys Thr Met Leu Ala Ile Ala Gly His Glu Gly Ala Leu Asn Glu
                165                 170                 175

Ile Glu Thr Ser Arg Leu Glu Gly Val Ile Asn Phe Glu Asn Leu Lys
            180                 185                 190

Val Lys Asp Val Asp Thr Thr Pro Arg Ile Asn Val Thr Ala Phe Ala
        195                 200                 205

Ser Asn Ala Thr Tyr Glu Glu Val Tyr Glu Thr Val Met Asn Lys Pro
    210                 215                 220

Tyr Thr Arg Tyr Pro Val Tyr Glu Gly Asp Ile Asp Asn Ile Ile Gly
225                 230                 235                 240

Val Phe His Ser Lys Tyr Leu Leu Ala Trp Ser Asn Lys Lys Glu Asn
                245                 250                 255

Gln Ile Thr Asn Tyr Ser Ala Lys Pro Leu Phe Val Asn Glu His Asn
            260                 265                 270

Lys Ala Glu Trp Val Leu Arg Lys Met Thr Ile Ser Arg Lys His Leu
        275                 280                 285

Ala Ile Val Leu Asp Glu Phe Gly Gly Thr Glu Ala Ile Val Ser His
    290                 295                 300
```

```
Glu Asp Leu Ile Glu Glu Leu Leu Gly Met Glu Ile Glu Asp Glu Met
305                 310                 315                 320

Asp Lys Lys Glu Lys Glu Lys Leu Ser Gln Gln Gln Ile Gln Phe Gln
                325                 330                 335

Gln Arg Lys Asn Arg Asn Val Ser Ile
            340                 345
```

<210> 20
<211> 133
<212> PRT
<213> Staphylococcus aureus

<400> 20

```
Met Asn Lys Gln Gln Lys Glu Phe Lys Ser Phe Tyr Ser Ile Arg Lys
  1               5                  10                  15

Ser Ser Leu Gly Val Ala Ser Val Ala Ile Ser Thr Leu Leu Leu Leu
             20                  25                  30

Met Ser Asn Gly Glu Ala Gln Ala Ala Ala Glu Glu Thr Gly Gly Thr
         35                  40                  45

Asn Thr Glu Ala Gln Pro Lys Thr Glu Ala Val Ala Ser Pro Thr Thr
     50                  55                  60

Thr Ser Glu Lys Ala Pro Glu Thr Lys Pro Val Ala Asn Ala Val Ser
 65                  70                  75                  80

Val Ser Asn Lys Glu Val Glu Ala Pro Thr Ser Glu Thr Lys Glu Ala
                85                  90                  95

Lys Glu Val Lys Glu Val Lys Ala Pro Lys Glu Thr Lys Glu Val Lys
            100                 105                 110

Pro Ala Ala Lys Ala Thr Asn Asn Thr Tyr Pro Ile Leu Asn Gln Glu
        115                 120                 125

Leu Ile Arg Ser Asp
    130
```

<210> 21
<211> 205
<212> PRT
<213> Staphylococcus aureus

<400> 21

```
Asp His Gly Ile Val Phe Asn Ala Ser Leu Pro Leu Tyr Lys Asp Ala
  1               5                  10                  15

Ile His Gln Lys Gly Ser Met Arg Ser Asn Asp Asn Gly Asp Asp Met
             20                  25                  30

Ser Met Met Val Gly Thr Val Leu Ser Gly Phe Glu Tyr Arg Ala Gln
         35                  40                  45

Lys Glu Lys Tyr Asp Asn Leu Tyr Lys Phe Phe Lys Glu Asn Glu Lys
     50                  55                  60

Lys Tyr Gln Tyr Thr Gly Phe Thr Lys Glu Ala Ile Asn Lys Thr Gln
```

```
 65                  70                  75                  80

Asn Val Gly Tyr Lys Asn Glu Tyr Phe Tyr Ile Thr Tyr Ser Ser Arg
                    85                  90                  95

Ser Leu Lys Glu Tyr Arg Lys Tyr Tyr Glu Pro Leu Ile Arg Lys Asn
               100                 105                 110

Asp Lys Glu Phe Lys Glu Gly Met Glu Arg Ala Arg Lys Glu Val Asn
           115                 120                 125

Tyr Ala Ala Asn Thr Asp Ala Val Ala Thr Leu Phe Ser Thr Lys Lys
       130                 135                 140

Asn Phe Thr Lys Asp Asn Thr Val Asp Asp Val Ile Glu Leu Ser Asp
145                 150                 155                 160

Lys Leu Tyr Asn Leu Lys Asn Lys Pro Asp Lys Ser Thr Ile Thr Ile
                165                 170                 175

Gln Ile Gly Lys Pro Thr Ile Asn Thr Lys Lys Ala Phe Tyr Asp Asp
            180                 185                 190

Asn Arg Pro Ile Glu Tyr Gly Val His Ser Lys Asp Glu
        195                 200                 205
```

<210> 22
<211> 510
<212> PRT
<213> Staphylococcus aureus

<400> 22

```
Asp His Tyr Val Ile Gln Tyr Phe Ser Gly Leu Ile Gly Gly Arg Gly
  1               5                  10                  15

Arg Arg Ala Asn Leu Tyr Gly Leu Phe Asn Lys Ala Ile Glu Phe Glu
            20                  25                  30

Asn Ser Ser Phe Arg Gly Leu Tyr Gln Phe Ile Arg Phe Ile Asp Glu
        35                  40                  45

Leu Ile Glu Arg Gly Lys Asp Phe Gly Glu Glu Asn Val Val Gly Pro
    50                  55                  60

Asn Asp Asn Val Val Arg Met Met Thr Ile His Ser Ser Lys Gly Leu
65                  70                  75                  80

Glu Phe Pro Phe Val Ile Tyr Ser Gly Leu Ser Lys Asp Phe Asn Lys
                85                  90                  95

Arg Asp Leu Lys Gln Pro Val Ile Leu Asn Gln Gln Phe Gly Leu Gly
            100                 105                 110

Met Asp Tyr Phe Asp Val Asp Lys Glu Met Ala Phe Pro Ser Leu Ala
        115                 120                 125

Ser Val Ala Tyr Arg Ala Val Ala Glu Lys Glu Leu Val Ser Glu Glu
    130                 135                 140

Met Arg Leu Val Tyr Val Ala Leu Thr Arg Ala Lys Glu Gln Leu Tyr
145                 150                 155                 160
```

```
Leu Ile Gly Arg Val Lys Asn Asp Lys Ser Leu Leu Glu Leu Glu Gln
                165                 170                 175

Leu Ser Ile Ser Gly Glu His Ile Ala Val Asn Glu Arg Leu Thr Ser
            180                 185                 190

Pro Asn Pro Phe His Leu Ile Tyr Ser Ile Leu Ser Lys His Gln Ser
        195                 200                 205

Ala Ser Ile Pro Asp Asp Leu Lys Phe Glu Lys Asp Ile Ala Gln Ile
    210                 215                 220

Glu Asp Ser Ser Arg Pro Asn Val Asn Ile Ser Ile Val Tyr Phe Glu
225                 230                 235                 240

Asp Val Ser Thr Glu Thr Ile Leu Asp Asn Asp Glu Tyr Arg Ser Val
                245                 250                 255

Asn Gln Leu Glu Thr Met Gln Asn Gly Asn Glu Asp Val Lys Ala Gln
            260                 265                 270

Ile Lys His Gln Leu Asp Tyr Arg Tyr Pro Tyr Val Asn Asp Thr Lys
        275                 280                 285

Lys Pro Ser Lys Gln Ser Val Ser Glu Leu Lys Arg Gln Tyr Glu Thr
    290                 295                 300

Glu Glu Ser Gly Thr Ser Tyr Glu Arg Val Arg Gln Tyr Arg Ile Gly
305                 310                 315                 320

Phe Ser Thr Tyr Glu Arg Pro Lys Phe Leu Ser Glu Gln Gly Lys Arg
                325                 330                 335

Lys Ala Asn Glu Ile Gly Thr Leu Met His Thr Val Met Gln His Leu
            340                 345                 350

Pro Phe Lys Lys Glu Arg Ile Ser Glu Val Glu Leu His Gln Tyr Ile
        355                 360                 365

Asp Gly Leu Ile Asp Lys His Ile Ile Glu Ala Asp Ala Lys Lys Asp
    370                 375                 380

Ile Arg Met Asp Glu Ile Met Thr Phe Ile Asn Ser Glu Leu Tyr Ser
385                 390                 395                 400

Ile Ile Ala Glu Ala Glu Gln Val Tyr Arg Glu Leu Pro Phe Val Val
                405                 410                 415

Asn Gln Ala Leu Val Asp Gln Leu Pro Gln Gly Asp Glu Asp Val Ser
            420                 425                 430

Ile Ile Gln Gly Met Ile Asp Leu Ile Phe Val Lys Asp Gly Val His
        435                 440                 445

Tyr Phe Val Asp Tyr Lys Thr Asp Ala Phe Asn Arg Arg Arg Gly Met
    450                 455                 460

Thr Asp Glu Glu Ile Gly Thr Gln Leu Lys Asn Lys Tyr Lys Ile Gln
465                 470                 475                 480

Met Lys Tyr Tyr Gln Asn Thr Leu Gln Thr Ile Leu Asn Lys Glu Val
                485                 490                 495
```

Lys Gly Tyr Leu Tyr Phe Phe Lys Phe Gly Thr Leu Gln Leu
500 505 510

<210> 23
<211> 124
<212> PRT
<213> Staphylococcus aureus

<400> 23
Met Lys Phe Leu Ser Phe Lys Tyr Asn Asp Lys Thr Ser Tyr Gly Val
1 5 10 15

Lys Val Lys Arg Glu Asp Ala Val Trp Asp Leu Thr Gln Val Phe Ala
20 25 30

Asp Phe Ala Glu Gly Asp Phe His Pro Lys Thr Leu Leu Ala Gly Leu
35 40 45

Gln Gln Asn His Thr Leu Asp Phe Gln Glu Gln Val Arg Lys Ala Val
50 55 60

Val Ala Ala Glu Asp Ser Gly Lys Ala Glu Asp Tyr Lys Ile Ser Phe
65 70 75 80

Asn Asp Ile Glu Phe Leu Pro Pro Val Thr Pro Pro Asn Asn Val Ile
85 90 95

Ala Phe Gly Arg Asn Tyr Lys Asp His Ala Asn Glu Leu Asn His Glu
100 105 110

Val Glu Lys Leu Tyr Val Phe Thr Lys Ala Ala Ser
115 120

<210> 24
<211> 180
<212> PRT
<213> Staphylococcus aureus

<400> 24
Ser Gly Thr Gly Phe Ile Val Gly Lys Asn Thr Ile Val Thr Asn Lys
1 5 10 15

His Val Val Ala Gly Met Glu Ile Gly Ala His Ile Ile Ala His Pro
20 25 30

Asn Gly Glu Tyr Asn Asn Gly Gly Phe Tyr Lys Val Lys Lys Ile Val
35 40 45

Arg Tyr Ser Gly Gln Glu Asp Ile Ala Ile Leu His Val Glu Asp Lys
50 55 60

Ala Val His Pro Lys Asn Arg Asn Phe Lys Asp Tyr Thr Gly Ile Leu
65 70 75 80

Lys Ile Ala Ser Glu Ala Lys Glu Asn Glu Arg Ile Ser Ile Val Gly
85 90 95

Tyr Pro Glu Pro Tyr Ile Asn Lys Phe Gln Met Tyr Glu Ser Thr Gly
100 105 110

Lys Val Leu Ser Val Lys Gly Asn Met Ile Ile Thr Asp Ala Phe Val

115 120 125

Glu Pro Gly Asn Ser Gly Ser Ala Val Phe Asn Ser Lys Tyr Glu Val
130 135 140

Val Gly Val His Phe Gly Gly Asn Gly Pro Gly Asn Lys Ser Thr Lys
145 150 155 160

Gly Tyr Gly Val Tyr Phe Ser Pro Glu Ile Lys Lys Phe Ile Ala Asp
165 170 175

Asn Thr Asp Lys
180

<210> 25
<211> 239
<212> PRT
<213> Staphylococcus aureus

<400> 25
Met Asn Lys Asn Ile Ile Ile Lys Ser Ile Ala Ala Leu Thr Ile Leu
1 5 10 15

Thr Ser Ile Thr Gly Val Gly Thr Thr Met Val Glu Gly Ile Gln Gln
20 25 30

Thr Ala Lys Ala Glu Asn Thr Val Lys Gln Ile Thr Asn Thr Asn Val
35 40 45

Ala Pro Tyr Ser Gly Val Thr Trp Met Gly Ala Gly Thr Gly Phe Val
50 55 60

Val Gly Asn His Thr Ile Ile Thr Asn Lys His Val Thr Tyr His Met
65 70 75 80

Lys Val Gly Asp Glu Ile Lys Ala His Pro Asn Gly Phe Tyr Asn Asn
85 90 95

Gly Gly Gly Leu Tyr Lys Val Thr Lys Ile Val Asp Tyr Pro Gly Lys
100 105 110

Glu Asp Ile Ala Val Val Gln Val Glu Glu Lys Ser Thr Gln Pro Lys
115 120 125

Gly Arg Lys Phe Lys Asp Phe Thr Ser Lys Phe Asn Ile Ala Ser Glu
130 135 140

Ala Lys Glu Asn Glu Pro Ile Ser Val Ile Gly Tyr Pro Asn Pro Asn
145 150 155 160

Gly Asn Lys Leu Gln Met Tyr Glu Ser Thr Gly Lys Val Leu Ser Val
165 170 175

Asn Gly Asn Ile Val Ser Ser Asp Ala Ile Ile Gln Pro Gly Ser Ser
180 185 190

Gly Ser Pro Ile Leu Asn Ser Lys His Glu Ala Ile Gly Val Ile Tyr
195 200 205

Ala Gly Asn Lys Pro Ser Gly Glu Ser Thr Arg Gly Phe Ala Val Tyr
210 215 220

```
Phe Ser Pro Glu Ile Lys Lys Phe Ile Ala Asp Asn Leu Asp Lys
225                 230                 235

<210> 26
<211> 470
<212> PRT
<213> Staphylococcus aureus

<400> 26
Met Gly Cys Thr Val Lys Met Asn Lys Ile Asn Asp Arg Asp Leu Thr
  1               5                  10                  15

Glu Leu Ser Ser Tyr Trp Val Tyr Gln Asn Ile Asp Ile Lys Lys Glu
             20                  25                  30

Phe Lys Val Asn Gly Lys Arg Phe Lys Gln Val Asp Ser Tyr Asn Asp
         35                  40                  45

Asp Lys Asn Ser Asn Leu Asn Gly Ala Ala Asp Ile Lys Ile Tyr Glu
     50                  55                  60

Leu Leu Asp Asp Lys Ser Lys Pro Thr Gly Gln Gln Thr Ile Ile Tyr
 65                  70                  75                  80

Gln Gly Thr Ser Asn Glu Ala Ile Asn Pro Asn Asn Pro Leu Lys Ser
                 85                  90                  95

Ser Gly Phe Gly Asp Asp Trp Leu Gln Asn Ala Lys Leu Met Asn Asn
            100                 105                 110

Asp Asn Glu Ser Thr Asp Tyr Leu Lys Gln Thr Asp Gln Leu Ser Asn
        115                 120                 125

Gln Tyr Lys Ile Lys Leu Glu Asp Ala Asp Arg Leu Ser Asn Ser Asp
    130                 135                 140

Phe Leu Lys Lys Tyr Arg Met Glu Ser Ser Asn Phe Lys Asn Lys Thr
145                 150                 155                 160

Ile Val Ala Asp Gly Gly Asn Ser Glu Gly Gly Ala Gly Ala Lys Tyr
                165                 170                 175

Gln Gly Ala Lys His Pro Asn Glu Lys Val Val Ala Thr Asp Ser Ala
            180                 185                 190

Met Ile Pro Tyr Ala Ala Trp Gln Lys Phe Ala Arg Pro Arg Phe Asp
        195                 200                 205

Asn Met Ile Ser Phe Asn Ser Thr Asn Asp Leu Leu Thr Trp Leu Gln
    210                 215                 220

Asp Pro Phe Ile Lys Asp Met Pro Gly Lys Arg Val Asn Ile Asn Asp
225                 230                 235                 240

Gly Val Pro Arg Leu Asp Thr Leu Ile Asp Ser His Val Gly Tyr Lys
                245                 250                 255

Arg Lys Leu Asn Arg Lys Asp Asn Thr Tyr Asp Thr Val Pro Leu Ile
            260                 265                 270

Lys Ile Lys Ser Val Lys Asp Thr Glu Ile Lys Asn Gly Lys Lys Val
        275                 280                 285
```

```
Lys Lys Thr Ile Asn Ile Thr Leu Asp Met Asp Gly Arg Ile Pro Ile
    290                 295                 300

Asn Val Trp Thr Gly Asp Ser Ile Ala Arg Ser Gly Arg Gly Thr Leu
305                 310                 315                 320

Ile Lys Leu Asn Leu Glu Asn Leu Asp Ala Leu Ser Lys Leu Ile Thr
                325                 330                 335

Gly Glu Thr Ser Gly Met Leu Ala Glu Cys Val Ile Phe Leu Asn Glu
            340                 345                 350

Ser Phe Asn Ile Ser Glu Asn Glu Asn Lys Asn Phe Ala Asp Arg Lys
        355                 360                 365

Lys Gln Leu Ser Glu Gly Phe Lys Asp Lys Ile Asn Leu Phe Gln Leu
    370                 375                 380

Glu Glu Met Glu Arg Thr Leu Ile Ser Lys Ile Asn Ser Leu Glu Glu
385                 390                 395                 400

Val Ala Asp Glu Thr Ile Glu Ser Ile Ser Ala Val Lys His Leu Leu
                405                 410                 415

Pro Asp Phe Ala Leu Asp Ala Leu Lys Glu Arg Ile Asn Glu Leu Phe
            420                 425                 430

Lys Gly Ile Lys Ser Phe Ile Glu Lys Val Tyr Asp Ser Ile Asp Asn
        435                 440                 445

Glu Ile Leu Glu Ile Phe Lys Asn Ile Asp His Asp Phe Arg Asp Gly
    450                 455                 460

Val Ser Glu Glu Met Met
465                 470


<210> 27
<211> 306
<212> PRT
<213> Staphylococcus aureus

<400> 27
Met Lys Lys Lys Asp Gly Thr Gln Gln Phe Tyr His Tyr Ala Ser Ser
  1               5                  10                  15

Val Lys Pro Ala Arg Val Ile Phe Thr Asp Ser Lys Pro Glu Ile Glu
            20                  25                  30

Leu Gly Leu Gln Ser Gly Gln Phe Trp Arg Lys Phe Glu Val Tyr Glu
        35                  40                  45

Gly Asp Lys Lys Leu Pro Ile Lys Leu Val Ser Tyr Asp Thr Val Lys
    50                  55                  60

Asp Tyr Ala Tyr Ile Arg Phe Ser Val Ser Asn Gly Thr Lys Ala Val
65                  70                  75                  80

Lys Ile Val Ser Ser Thr His Phe Asn Asn Lys Glu Glu Lys Tyr Asp
                85                  90                  95

Tyr Thr Leu Met Glu Phe Ala Gln Pro Ile Tyr Asn Ser Ala Asp Lys
```

100 105 110

Phe Lys Thr Glu Glu Asp Tyr Lys Ala Glu Lys Leu Leu Ala Pro Tyr
115 120 125

Lys Lys Ala Lys Thr Leu Glu Arg Gln Val Tyr Glu Leu Asn Lys Ile
130 135 140

Gln Asp Lys Leu Pro Glu Lys Leu Lys Ala Glu Tyr Lys Lys Lys Leu
145 150 155 160

Glu Asp Thr Lys Lys Ala Leu Asp Glu Gln Val Lys Ser Ala Ile Thr
165 170 175

Glu Phe Gln Asn Val Gln Pro Thr Asn Glu Lys Met Thr Asp Leu Gln
180 185 190

Asp Thr Lys Tyr Val Val Tyr Glu Ser Val Glu Asn Asn Glu Ser Met
195 200 205

Met Asp Thr Phe Val Lys His Pro Ile Lys Thr Gly Met Leu Asn Gly
210 215 220

Lys Lys Tyr Met Val Met Glu Thr Thr Asn Asp Asp Tyr Trp Lys Asp
225 230 235 240

Phe Met Val Glu Gly Gln Arg Val Arg Thr Ile Ser Lys Asp Ala Lys
245 250 255

Asn Asn Thr Arg Thr Ile Ile Phe Pro Tyr Val Glu Gly Lys Thr Leu
260 265 270

Tyr Asp Ala Ile Val Lys Val His Val Lys Thr Ile Asp Tyr Asp Gly
275 280 285

Gln Tyr His Val Arg Ile Val Asp Lys Glu Ala Phe Thr Lys Ala His
290 295 300

Thr Asp
305

<210> 28
<211> 2659
<212> PRT
<213> Staphylococcus aureus

<400> 28
Asp Gln Thr Thr Ile Ile Asn Ser Leu Thr Phe Thr Glu Thr Val Pro
1 5 10 15

Asn Arg Ser Tyr Ala Arg Ala Ser Ala Asn Glu Ile Thr Ser Lys Thr
20 25 30

Val Ser Asn Val Ser Arg Thr Gly Asn Asn Ala Asn Val Thr Val Thr
35 40 45

Val Thr Tyr Gln Asp Gly Thr Thr Ser Thr Val Thr Val Pro Val Lys
50 55 60

His Val Ile Pro Glu Ile Val Ala His Ser His Tyr Thr Val Gln Gly
65 70 75 80

```
Gln Asp Phe Pro Ala Gly Asn Gly Ser Ser Ala Ser Asp Tyr Phe Lys
                  85                  90                  95

Leu Ser Asn Gly Ser Asp Ile Ala Asp Ala Thr Ile Thr Trp Val Ser
             100                 105                 110

Gly Gln Ala Pro Asn Lys Asp Asn Thr Arg Ile Gly Glu Asp Ile Thr
         115                 120                 125

Val Thr Ala His Ile Leu Ile Asp Gly Glu Thr Thr Pro Ile Thr Lys
     130                 135                 140

Thr Ala Thr Tyr Lys Val Val Arg Thr Val Pro Lys His Val Phe Glu
145                 150                 155                 160

Thr Ala Arg Gly Val Leu Tyr Pro Gly Val Ser Asp Met Tyr Asp Ala
                 165                 170                 175

Lys Gln Tyr Val Lys Pro Val Asn Asn Ser Trp Ser Thr Asn Ala Gln
             180                 185                 190

His Met Asn Phe Gln Phe Val Gly Thr Tyr Gly Pro Asn Lys Asp Val
         195                 200                 205

Val Gly Ile Ser Thr Arg Leu Ile Arg Val Thr Tyr Asp Asn Arg Gln
     210                 215                 220

Thr Glu Asp Leu Thr Ile Leu Ser Lys Val Lys Pro Asp Pro Pro Arg
225                 230                 235                 240

Ile Asp Ala Asn Ser Val Thr Tyr Lys Ala Gly Leu Thr Asn Gln Glu
                 245                 250                 255

Ile Lys Val Asn Asn Val Leu Asn Asn Ser Ser Val Lys Leu Phe Lys
             260                 265                 270

Ala Asp Asn Thr Pro Leu Asn Val Thr Asn Ile Thr His Gly Ser Gly
         275                 280                 285

Phe Ser Ser Val Val Thr Val Ser Asp Ala Leu Pro Asn Gly Gly Ile
     290                 295                 300

Lys Ala Lys Ser Ser Ile Ser Met Asn Asn Val Thr Tyr Thr Thr Gln
305                 310                 315                 320

Asp Glu His Gly Gln Val Val Thr Val Thr Arg Asn Glu Ser Val Asp
                 325                 330                 335

Ser Asn Asp Ser Ala Thr Val Thr Val Thr Pro Gln Leu Gln Ala Thr
             340                 345                 350

Thr Glu Gly Ala Val Phe Ile Lys Gly Gly Asp Gly Phe Asp Phe Gly
         355                 360                 365

His Val Glu Arg Phe Ile Gln Asn Pro Pro His Gly Ala Thr Val Ala
     370                 375                 380

Trp His Asp Ser Pro Asp Thr Trp Lys Asn Thr Val Gly Asn Thr His
385                 390                 395                 400

Lys Thr Ala Val Val Thr Leu Pro Asn Gly Gln Gly Thr Arg Asn Val
                 405                 410                 415
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Glu Val Pro Val Lys Val Tyr Pro Val Ala Asn Ala Lys Ala Pro Ser
            420                 425                 430

Arg Asp Val Lys Gly Gln Asn Leu Thr Asn Gly Thr Asp Ala Met Asn
        435                 440                 445

Tyr Ile Thr Phe Asp Pro Asn Thr Asn Thr Asn Gly Ile Thr Ala Ala
    450                 455                 460

Trp Ala Asn Arg Gln Gln Pro Asn Asn Gln Gln Ala Gly Val Gln His
465                 470                 475                 480

Leu Asn Val Asp Val Thr Tyr Pro Gly Ile Ser Ala Ala Lys Arg Val
                485                 490                 495

Pro Val Thr Val Asn Val Tyr Gln Phe Glu Phe Pro Gln Thr Thr Tyr
            500                 505                 510

Thr Thr Thr Val Gly Gly Thr Leu Ala Ser Gly Thr Gln Ala Ser Gly
        515                 520                 525

Tyr Ala His Met Gln Asn Ala Thr Gly Leu Pro Thr Asp Gly Phe Thr
    530                 535                 540

Tyr Lys Trp Asn Arg Asp Thr Thr Gly Thr Asn Asp Ala Asn Trp Ser
545                 550                 555                 560

Ala Met Asn Lys Pro Asn Val Ala Lys Val Val Asn Ala Lys Tyr Asp
                565                 570                 575

Val Ile Tyr Asn Gly His Thr Phe Ala Thr Ser Leu Pro Ala Lys Phe
            580                 585                 590

Val Val Lys Asp Val Gln Pro Ala Lys Pro Thr Val Thr Glu Thr Ala
        595                 600                 605

Ala Gly Ala Ile Thr Ile Ala Pro Gly Ala Asn Gln Thr Val Asn Thr
    610                 615                 620

His Ala Gly Asn Val Thr Thr Tyr Ala Asp Lys Leu Val Ile Lys Arg
625                 630                 635                 640

Asn Gly Asn Val Val Thr Thr Phe Thr Arg Arg Asn Asn Thr Ser Pro
                645                 650                 655

Trp Val Lys Glu Ala Ser Ala Ala Thr Val Ala Gly Ile Ala Gly Thr
            660                 665                 670

Asn Asn Gly Ile Thr Val Ala Ala Gly Thr Phe Asn Pro Ala Asp Thr
        675                 680                 685

Ile Gln Val Val Ala Thr Gln Gly Ser Gly Glu Thr Val Ser Asp Glu
    690                 695                 700

Gln Arg Ser Asp Asp Phe Thr Val Val Ala Pro Gln Pro Asn Gln Ala
705                 710                 715                 720

Thr Thr Lys Ile Trp Gln Asn Gly His Ile Asp Ile Thr Pro Asn Asn
                725                 730                 735

Pro Ser Gly His Leu Ile Asn Pro Thr Gln Ala Met Asp Ile Ala Tyr
            740                 745                 750
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```
Thr Glu Lys Val Gly Asn Gly Ala Glu His Ser Lys Thr Ile Asn Val
        755                 760                 765

Val Arg Gly Gln Asn Asn Gln Trp Thr Ile Ala Asn Lys Pro Asp Tyr
    770                 775                 780

Val Thr Leu Asp Ala Gln Thr Gly Lys Val Thr Phe Asn Ala Asn Thr
785                 790                 795                 800

Ile Lys Pro Asn Ser Ser Ile Thr Ile Thr Pro Lys Ala Gly Thr Gly
                805                 810                 815

His Ser Val Ser Ser Asn Pro Ser Thr Leu Thr Ala Pro Ala Ala His
            820                 825                 830

Thr Val Asn Thr Thr Glu Ile Val Lys Asp Tyr Gly Ser Asn Val Thr
        835                 840                 845

Ala Ala Glu Ile Asn Asn Ala Val Gln Val Ala Asn Lys Arg Thr Ala
    850                 855                 860

Thr Ile Lys Asn Gly Thr Ala Met Pro Thr Asn Leu Ala Gly Gly Ser
865                 870                 875                 880

Thr Thr Thr Ile Pro Val Thr Val Thr Tyr Asn Asp Gly Ser Thr Glu
                885                 890                 895

Glu Val Gln Glu Ser Ile Phe Thr Lys Ala Asp Lys Arg Glu Leu Ile
            900                 905                 910

Thr Ala Lys Asn His Leu Asp Asp Pro Val Ser Thr Glu Gly Lys Lys
        915                 920                 925

Pro Gly Thr Ile Thr Gln Tyr Asn Asn Ala Met His Asn Ala Gln Gln
    930                 935                 940

Gln Ile Asn Thr Ala Lys Thr Glu Ala Gln Gln Val Ile Asn Asn Glu
945                 950                 955                 960

Arg Ala Thr Pro Gln Gln Val Ser Asp Ala Leu Thr Lys Val Arg Ala
                965                 970                 975

Ala Gln Thr Lys Ile Asp Gln Ala Lys Ala Leu Leu Gln Asn Lys Glu
            980                 985                 990

Asp Asn Ser Gln Leu Val Thr Ser Lys Asn Asn Leu Gln Ser Ser Val
        995                1000                1005

Asn Gln Val Pro Ser Thr Ala Gly Met Thr Gln Gln Ser Ile Asp Asn
   1010                1015                1020

Tyr Asn Ala Lys Lys Arg Glu Ala Glu Thr Glu Ile Thr Ala Ala Gln
1025                1030                1035                1040

Arg Val Ile Asp Asn Gly Asp Ala Thr Ala Gln Gln Ile Ser Asp Glu
                1045                1050                1055

Lys His Arg Val Asp Asn Ala Leu Thr Ala Leu Asn Gln Ala Lys His
            1060                1065                1070

Asp Leu Thr Ala Asp Thr His Ala Leu Glu Gln Ala Val Gln Gln Leu
        1075                1080                1085
```

```
Asn Arg Thr Gly Thr Thr Thr Gly Lys Lys Pro Ala Ser Ile Thr Ala
   1090                1095                1100

Tyr Asn Asn Ser Ile Arg Ala Leu Gln Ser Asp Leu Thr Ser Ala Lys
1105                1110                1115                1120

Asn Ser Ala Asn Ala Ile Ile Gln Lys Pro Ile Arg Thr Val Gln Glu
                1125                1130                1135

Val Gln Ser Ala Leu Thr Asn Val Asn Arg Val Asn Glu Arg Leu Thr
            1140                1145                1150

Gln Ala Ile Asn Gln Leu Val Pro Leu Ala Asp Asn Ser Ala Leu Lys
        1155                1160                1165

Thr Ala Lys Thr Lys Leu Asp Glu Glu Ile Asn Lys Ser Val Thr Thr
   1170                1175                1180

Asp Gly Met Thr Gln Ser Ser Ile Gln Ala Tyr Glu Asn Ala Lys Arg
1185                1190                1195                1200

Ala Gly Gln Thr Glu Ser Thr Asn Ala Gln Asn Val Ile Asn Asn Gly
                1205                1210                1215

Asp Ala Thr Asp Gln Gln Ile Ala Ala Glu Lys Thr Lys Val Glu Glu
            1220                1225                1230

Lys Tyr Asn Ser Leu Lys Gln Ala Ile Ala Gly Leu Thr Pro Asp Leu
        1235                1240                1245

Ala Pro Leu Gln Thr Ala Lys Thr Gln Leu Gln Asn Asp Ile Asp Gln
   1250                1255                1260

Pro Thr Ser Thr Thr Gly Met Thr Ser Ala Ser Ile Ala Ala Phe Asn
1265                1270                1275                1280

Glu Lys Leu Ser Ala Ala Arg Thr Lys Ile Gln Glu Ile Asp Arg Val
                1285                1290                1295

Leu Ala Ser His Pro Asp Val Ala Thr Ile Arg Gln Asn Val Thr Ala
            1300                1305                1310

Ala Asn Ala Ala Lys Ser Ala Leu Asp Gln Ala Arg Asn Gly Leu Thr
        1315                1320                1325

Val Asp Lys Ala Pro Leu Glu Asn Ala Lys Asn Gln Leu Gln Tyr Ser
   1330                1335                1340

Ile Asp Thr Gln Thr Ser Thr Thr Gly Met Thr Gln Asp Ser Ile Asn
1345                1350                1355                1360

Ala Tyr Asn Ala Lys Leu Thr Ala Ala Arg Asn Lys Ile Gln Gln Ile
                1365                1370                1375

Asn Gln Val Leu Ala Gly Ser Pro Thr Val Glu Gln Ile Asn Thr Asn
            1380                1385                1390

Thr Ser Thr Ala Asn Gln Ala Lys Ser Asp Leu Asp His Ala Arg Gln
        1395                1400                1405

Ala Leu Thr Pro Asp Lys Ala Pro Leu Gln Thr Ala Lys Thr Gln Leu
   1410                1415                1420
```

```
Glu Gln Ser Ile Asn Gln Pro Thr Asp Thr Thr Gly Met Thr Thr Ala
1425                1430                1435                1440

Ser Leu Asn Ala Tyr Asn Gln Lys Leu Gln Ala Ala Arg Gln Lys Leu
                1445                1450                1455

Thr Glu Ile Asn Gln Val Leu Asn Gly Asn Pro Thr Val Gln Asn Ile
            1460                1465                1470

Asn Asp Lys Val Thr Glu Ala Asn Gln Ala Lys Asp Gln Leu Asn Thr
        1475                1480                1485

Ala Arg Gln Gly Leu Thr Leu Asp Arg Gln Pro Ala Leu Thr Thr Leu
    1490                1495                1500

His Gly Ala Ser Asn Leu Asn Gln Ala Gln Gln Asn Asn Phe Thr Gln
1505                1510                1515                1520

Gln Ile Asn Ala Ala Gln Asn His Ala Ala Leu Glu Thr Ile Lys Ser
                1525                1530                1535

Asn Ile Thr Ala Leu Asn Thr Ala Met Thr Lys Leu Lys Asp Ser Val
            1540                1545                1550

Ala Asp Asn Asn Thr Ile Lys Ser Asp Gln Asn Tyr Thr Asp Ala Thr
        1555                1560                1565

Pro Ala Asn Lys Gln Ala Tyr Asp Asn Ala Val Asn Ala Ala Lys Gly
    1570                1575                1580

Val Ile Gly Glu Thr Thr Asn Pro Thr Met Asp Val Asn Thr Val Asn
1585                1590                1595                1600

Gln Lys Ala Ala Ser Val Lys Ser Thr Lys Asp Ala Leu Asp Gly Gln
                1605                1610                1615

Gln Asn Leu Gln Arg Ala Lys Thr Glu Ala Thr Asn Ala Ile Thr His
            1620                1625                1630

Ala Ser Asp Leu Asn Gln Ala Gln Lys Asn Ala Leu Thr Gln Gln Val
        1635                1640                1645

Asn Ser Ala Gln Asn Val Gln Ala Val Asn Asp Ile Lys Gln Thr Thr
    1650                1655                1660

Gln Ser Leu Asn Thr Ala Met Thr Gly Leu Lys Arg Gly Val Ala Asn
1665                1670                1675                1680

His Asn Gln Val Val Gln Ser Asp Asn Tyr Val Asn Ala Asp Thr Asn
                1685                1690                1695

Lys Lys Asn Asp Tyr Asn Asn Ala Tyr Asn His Ala Asn Asp Ile Ile
            1700                1705                1710

Asn Gly Asn Ala Gln His Pro Val Ile Thr Pro Ser Asp Val Asn Asn
        1715                1720                1725

Ala Leu Ser Asn Val Thr Ser Lys Glu His Ala Leu Asn Gly Glu Ala
    1730                1735                1740

Lys Leu Asn Ala Ala Lys Gln Glu Ala Asn Thr Ala Leu Gly His Leu
1745                1750                1755                1760
```

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Asn Asn Leu Asn Asn Ala Gln Arg Gln Asn Leu Gln Ser Gln Ile Asn
                1765                1770                1775

Gly Ala His Gln Ile Asp Ala Val Asn Thr Ile Lys Gln Asn Ala Thr
            1780                1785                1790

Asn Leu Asn Ser Ala Met Gly Asn Leu Arg Gln Ala Val Ala Asp Lys
        1795                1800                1805

Asp Gln Val Lys Arg Thr Glu Asp Tyr Ala Asp Ala Asp Thr Ala Lys
    1810                1815                1820

Gln Asn Ala Tyr Asn Ser Ala Val Ser Ser Ala Glu Thr Ile Ile Asn
1825                1830                1835                1840

Gln Thr Thr Asn Pro Thr Met Ser Val Asp Asp Val Asn Arg Ala Thr
                1845                1850                1855

Ser Ala Val Thr Ser Asn Lys Asn Ala Leu Asn Gly Tyr Glu Lys Leu
            1860                1865                1870

Ala Gln Ser Lys Thr Asp Ala Ala Arg Ala Ile Asp Ala Leu Pro His
        1875                1880                1885

Leu Asn Asn Ala Gln Lys Ala Asp Val Lys Ser Lys Ile Asn Ala Ala
    1890                1895                1900

Ser Asn Ile Ala Gly Val Asn Thr Val Lys Gln Gln Gly Thr Asp Leu
1905                1910                1915                1920

Asn Thr Ala Met Gly Asn Leu Gln Gly Ala Ile Asn Asp Glu Gln Thr
                1925                1930                1935

Thr Leu Asn Ser Gln Asn Tyr Gln Asp Ala Thr Pro Ser Lys Lys Thr
            1940                1945                1950

Ala Tyr Thr Asn Ala Val Gln Ala Ala Lys Asp Ile Leu Asn Lys Ser
        1955                1960                1965

Asn Gly Gln Asn Lys Thr Lys Asp Gln Val Thr Glu Ala Met Asn Gln
    1970                1975                1980

Val Asn Ser Ala Lys Asn Asn Leu Asp Gly Thr Arg Leu Leu Asp Gln
1985                1990                1995                2000

Ala Lys Gln Thr Ala Lys Gln Gln Leu Asn Asn Met Thr His Leu Thr
                2005                2010                2015

Thr Ala Gln Lys Thr Asn Leu Thr Asn Gln Ile Asn Ser Gly Thr Thr
            2020                2025                2030

Val Ala Gly Val Gln Thr Val Gln Ser Asn Ala Asn Thr Leu Asp Gln
        2035                2040                2045

Ala Met Asn Thr Leu Arg Gln Ser Ile Ala Asn Lys Asp Ala Thr Lys
    2050                2055                2060

Ala Ser Glu Asp Tyr Val Asp Ala Asn Asn Asp Lys Gln Thr Ala Tyr
2065                2070                2075                2080

Asn Asn Ala Val Ala Ala Ala Glu Thr Ile Ile Asn Ala Asn Ser Asn
                2085                2090                2095
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Pro Glu Met Asn Pro Ser Thr Ile Thr Gln Lys Ala Glu Gln Val Asn
            2100                2105                2110

Ser Ser Lys Thr Ala Leu Asn Gly Asp Glu Asn Leu Ala Ala Ala Lys
        2115                2120                2125

Gln Asn Ala Lys Thr Tyr Leu Asn Thr Leu Thr Ser Ile Thr Asp Ala
    2130                2135                2140

Gln Lys Asn Asn Leu Ile Ser Gln Ile Thr Ser Ala Thr Arg Val Ser
2145                2150                2155                2160

Gly Val Asp Thr Val Lys Gln Asn Ala Gln His Leu Asp Gln Ala Met
                2165                2170                2175

Ala Ser Leu Gln Asn Gly Ile Asn Asn Glu Ser Gln Val Lys Ser Ser
            2180                2185                2190

Glu Lys Tyr Arg Asp Ala Asp Thr Asn Lys Gln Gln Glu Tyr Asp Asn
        2195                2200                2205

Ala Ile Thr Ala Ala Lys Ala Ile Leu Asn Lys Ser Thr Gly Pro Asn
    2210                2215                2220

Thr Ala Gln Asn Ala Val Glu Ala Ala Leu Gln Arg Val Asn Asn Ala
2225                2230                2235                2240

Lys Asp Ala Leu Asn Gly Asp Ala Lys Leu Ile Ala Ala Gln Asn Ala
                2245                2250                2255

Ala Lys Gln His Leu Gly Thr Leu Thr His Ile Thr Thr Ala Gln Arg
            2260                2265                2270

Asn Asp Leu Thr Asn Gln Ile Ser Gln Ala Thr Asn Leu Ala Gly Val
        2275                2280                2285

Glu Ser Val Lys Gln Asn Ala Asn Ser Leu Asp Gly Ala Met Gly Asn
    2290                2295                2300

Leu Gln Thr Ala Ile Asn Asp Lys Ser Gly Thr Leu Ala Ser Gln Asn
2305                2310                2315                2320

Phe Leu Asp Ala Asp Glu Gln Lys Arg Asn Ala Tyr Asn Gln Ala Val
                2325                2330                2335

Ser Ala Ala Glu Thr Ile Leu Asn Lys Gln Thr Gly Pro Asn Thr Ala
            2340                2345                2350

Lys Thr Ala Val Glu Gln Ala Leu Asn Asn Val Asn Asn Ala Lys His
        2355                2360                2365

Ala Leu Asn Gly Thr Gln Asn Leu Asn Asn Ala Lys Gln Ala Ala Ile
    2370                2375                2380

Thr Ala Ile Asn Gly Ala Ser Asp Leu Asn Gln Lys Gln Lys Asp Ala
2385                2390                2395                2400

Leu Lys Ala Gln Ala Asn Gly Ala Gln Arg Val Ser Asn Ala Gln Asp
                2405                2410                2415

Val Gln His Asn Ala Thr Glu Leu Asn Thr Ala Met Gly Thr Leu Lys
            2420                2425                2430
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```
His Ala Ile Ala Asp Lys Thr Asn Thr Leu Ala Ser Ser Lys Tyr Val
      2435                2440                2445

Asn Ala Asp Ser Thr Lys Gln Asn Ala Tyr Thr Thr Lys Val Thr Asn
   2450                2455                2460

Ala Glu His Ile Ile Ser Gly Thr Pro Thr Val Val Thr Thr Pro Ser
2465                2470                2475                2480

Glu Val Thr Ala Ala Ala Asn Gln Val Asn Ser Ala Lys Gln Glu Leu
                2485                2490                2495

Asn Gly Asp Glu Arg Leu Arg Glu Ala Lys Gln Asn Ala Asn Thr Ala
            2500                2505                2510

Ile Asp Ala Leu Thr Gln Leu Asn Thr Pro Gln Lys Ala Lys Leu Lys
      2515                2520                2525

Glu Gln Val Gly Gln Ala Asn Arg Leu Glu Asp Val Gln Thr Val Gln
   2530                2535                2540

Thr Asn Gly Gln Ala Leu Asn Asn Ala Met Lys Gly Leu Arg Asp Ser
2545                2550                2555                2560

Ile Ala Asn Glu Thr Thr Val Lys Thr Ser Gln Asn Tyr Thr Asp Ala
                2565                2570                2575

Ser Pro Asn Asn Gln Ser Thr Tyr Asn Ser Ala Val Ser Asn Ala Lys
            2580                2585                2590

Gly Ile Ile Asn Gln Thr Asn Asn Pro Thr Met Asp Thr Ser Ala Ile
      2595                2600                2605

Thr Gln Ala Thr Thr Gln Val Asn Asn Ala Lys Asn Gly Leu Asn Gly
   2610                2615                2620

Ala Glu Asn Leu Arg Asn Ala Gln Asn Thr Ala Lys Gln Asn Leu Asn
2625                2630                2635                2640

Thr Leu Ser His Leu Thr Asn Asn Gln Lys Ser Ala Ile Ser Ser Gln
                2645                2650                2655

Ile Asp Arg
```

<210> 29
<211> 496
<212> PRT
<213> Staphylococcus aureus

<400> 29

```
Met Asn Met Lys Lys Lys Glu Lys His Ala Ile Arg Lys Lys Ser Ile
  1               5                  10                  15

Gly Val Ala Ser Val Leu Val Gly Thr Leu Ile Gly Phe Gly Leu Leu
            20                  25                  30

Ser Ser Lys Glu Ala Asp Ala Ser Glu Asn Ser Val Thr Gln Ser Asp
        35                  40                  45

Ser Ala Ser Asn Glu Ser Lys Ser Asn Asp Ser Ser Ser Val Ser Ala
    50                  55                  60
```

```
Ala Pro Lys Thr Asp Asp Thr Asn Val Ser Asp Thr Lys Thr Ser Ser
 65                  70                  75                  80

Asn Thr Asn Asn Gly Glu Thr Ser Val Ala Gln Asn Pro Ala Gln Gln
                 85                  90                  95

Glu Thr Thr Gln Ser Ser Ser Thr Asn Ala Thr Thr Glu Glu Thr Pro
            100                 105                 110

Val Thr Gly Glu Ala Thr Thr Thr Thr Thr Asn Gln Ala Asn Thr Pro
        115                 120                 125

Ala Thr Thr Gln Ser Ser Asn Thr Asn Ala Glu Glu Leu Val Asn Gln
    130                 135                 140

Thr Ser Asn Glu Thr Thr Phe Asn Asp Thr Asn Thr Val Ser Ser Val
145                 150                 155                 160

Asn Ser Pro Gln Asn Ser Thr Asn Ala Glu Asn Val Ser Thr Thr Gln
                165                 170                 175

Asp Thr Ser Thr Glu Ala Thr Pro Ser Asn Asn Glu Ser Ala Pro Gln
            180                 185                 190

Ser Thr Asp Ala Ser Asn Lys Asp Val Val Asn Gln Ala Val Asn Thr
        195                 200                 205

Ser Ala Pro Arg Met Arg Ala Phe Ser Leu Ala Ala Val Ala Ala Asp
    210                 215                 220

Ala Pro Ala Ala Gly Thr Asp Ile Thr Asn Gln Leu Thr Asn Val Thr
225                 230                 235                 240

Val Gly Ile Asp Ser Gly Thr Thr Val Tyr Pro His Gln Ala Gly Tyr
                245                 250                 255

Val Lys Leu Asn Tyr Gly Phe Ser Val Pro Asn Ser Ala Val Lys Gly
            260                 265                 270

Asp Thr Phe Lys Ile Thr Val Pro Lys Glu Leu Asn Leu Asn Gly Val
        275                 280                 285

Thr Ser Thr Ala Lys Val Pro Pro Ile Met Ala Gly Asp Gln Val Leu
    290                 295                 300

Ala Asn Gly Val Ile Asp Ser Asp Gly Asn Val Ile Tyr Thr Phe Thr
305                 310                 315                 320

Asp Tyr Val Asn Thr Lys Asp Asp Val Lys Ala Thr Leu Thr Met Pro
                325                 330                 335

Ala Tyr Ile Asp Pro Glu Asn Val Lys Lys Thr Gly Asn Val Thr Leu
            340                 345                 350

Ala Thr Gly Ile Gly Ser Thr Thr Ala Asn Lys Thr Val Leu Val Asp
        355                 360                 365

Tyr Glu Lys Tyr Gly Lys Phe Tyr Asn Leu Ser Ile Lys Gly Thr Ile
    370                 375                 380

Asp Gln Ile Asp Lys Thr Asn Asn Thr Tyr Arg Gln Thr Ile Tyr Val
385                 390                 395                 400
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Asn Pro Ser Gly Asp Asn Val Ile Ala Pro Val Leu Thr Gly Asn Leu
405 410 415

Lys Pro Asn Thr Asp Ser Asn Ala Leu Ile Asp Gln Gln Asn Thr Ser
420 425 430

Ile Lys Val Tyr Lys Val Asp Asn Ala Ala Asp Leu Ser Glu Ser Tyr
435 440 445

Phe Val Asn Pro Glu Asn Phe Glu Asp Val Thr Asn Ser Val Asn Ile
450 455 460

Thr Phe Pro Asn Pro Asn Gln Tyr Lys Val Glu Phe Asn Thr Pro Asp
465 470 475 480

Asp Gln Ile Thr Thr Pro Tyr Ile Val Val Val Asn Gly His Ile Asp
485 490 495

<210> 30
<211> 541
<212> PRT
<213> Staphylococcus aureus

<400> 30
Asp Gln Tyr Leu Leu Glu Arg Lys Lys Ser Gln Tyr Glu Asp Tyr Lys
1 5 10 15

Gln Trp Tyr Ala Asn Tyr Lys Lys Glu Asn Pro Arg Thr Asp Leu Lys
20 25 30

Met Ala Asn Phe His Lys Tyr Asn Leu Glu Glu Leu Ser Met Lys Glu
35 40 45

Tyr Asn Glu Leu Gln Asp Ala Leu Lys Arg Ala Leu Asp Asp Phe His
50 55 60

Arg Glu Val Lys Asp Ile Lys Asp Lys Asn Ser Asp Leu Lys Thr Phe
65 70 75 80

Asn Ala Ala Glu Glu Asp Lys Ala Thr Lys Glu Val Tyr Asp Leu Val
85 90 95

Ser Glu Ile Asp Thr Leu Val Val Ser Tyr Tyr Gly Asp Lys Asp Tyr
100 105 110

Gly Glu His Ala Lys Glu Leu Arg Ala Lys Leu Asp Leu Ile Leu Gly
115 120 125

Asp Thr Asp Asn Pro His Lys Ile Thr Asn Glu Arg Ile Lys Lys Glu
130 135 140

Met Ile Asp Asp Leu Asn Ser Ile Ile Asp Asp Phe Phe Met Glu Thr
145 150 155 160

Lys Gln Asn Arg Pro Lys Ser Ile Thr Lys Tyr Asn Pro Thr Thr His
165 170 175

Asn Tyr Lys Thr Asn Ser Asp Asn Lys Pro Asn Phe Asp Lys Leu Val

180 185 190

Glu Glu Thr Lys Lys Ala Val Lys Glu Ala Asp Asp Ser Trp Lys Lys
195 200 205

Lys Thr Val Lys Lys Tyr Gly Glu Thr Glu Thr Lys Ser Pro Val Val
210 215 220

Lys Glu Glu Lys Lys Val Glu Glu Pro Gln Ala Pro Lys Val Asp Asn
225 230 235 240

Gln Gln Glu Val Lys Thr Thr Ala Gly Lys Ala Glu Glu Thr Thr Gln
245 250 255

Pro Val Ala Gln Pro Leu Val Lys Ile Pro Gln Gly Thr Ile Thr Gly
260 265 270

Glu Ile Val Lys Gly Pro Glu Tyr Pro Thr Met Glu Asn Lys Thr Val
275 280 285

Gln Gly Glu Ile Val Gln Gly Pro Asp Phe Leu Thr Met Glu Gln Ser
290 295 300

Gly Pro Ser Leu Ser Asn Asn Tyr Thr Asn Pro Pro Leu Thr Asn Pro
305 310 315 320

Ile Leu Glu Gly Leu Glu Gly Ser Ser Ser Lys Leu Glu Ile Lys Pro
325 330 335

Gln Gly Thr Glu Ser Thr Leu Lys Gly Thr Gln Gly Glu Ser Ser Asp
340 345 350

Ile Glu Val Lys Pro Gln Ala Thr Glu Thr Thr Glu Ala Ser Gln Tyr
355 360 365

Gly Pro Arg Pro Gln Phe Asn Lys Thr Pro Lys Tyr Val Lys Tyr Arg
370 375 380

Asp Ala Gly Thr Gly Ile Arg Glu Tyr Asn Asp Gly Thr Phe Gly Tyr
385 390 395 400

Glu Ala Arg Pro Arg Phe Asn Lys Pro Ser Glu Thr Asn Ala Tyr Asn
405 410 415

Val Thr Thr His Ala Asn Gly Gln Val Ser Tyr Gly Ala Arg Pro Thr
420 425 430

Tyr Lys Lys Pro Ser Glu Thr Asn Ala Tyr Asn Val Thr Thr His Ala
435 440 445

Asn Gly Gln Val Ser Tyr Gly Ala Arg Pro Thr Gln Asn Lys Pro Ser
450 455 460

Lys Thr Asn Ala Tyr Asn Val Thr Thr His Gly Asn Gly Gln Val Ser
465 470 475 480

Tyr Gly Ala Arg Gln Ala Gln Asn Lys Pro Ser Lys Thr Asn Ala Tyr
485 490 495

Asn Val Thr Thr His Ala Asn Gly Gln Val Ser Tyr Gly Ala Arg Pro
500 505 510

Thr Tyr Lys Lys Pro Ser Lys Thr Asn Ala Tyr Asn Val Thr Thr His

515 520 525

Ala Asp Gly Thr Ala Thr Tyr Gly Pro Arg Val Thr Lys
530 535 540

<210> 31
<211> 356
<212> PRT
<213> Staphylococcus aureus

<400> 31
Met Lys Met Arg Thr Ile Ala Lys Thr Ser Leu Ala Leu Gly Leu Leu
1 5 10 15

Thr Thr Gly Ala Ile Thr Val Thr Thr Gln Ser Val Lys Ala Glu Lys
20 25 30

Ile Gln Ser Thr Lys Val Asp Lys Val Pro Thr Leu Lys Ala Glu Arg
35 40 45

Leu Ala Met Ile Asn Ile Thr Ala Gly Ala Asn Ser Ala Thr Thr Gln
50 55 60

Ala Ala Asn Thr Arg Gln Glu Arg Thr Pro Lys Leu Glu Lys Ala Pro
65 70 75 80

Asn Thr Asn Glu Glu Lys Thr Ser Ala Ser Lys Ile Glu Lys Ile Ser
85 90 95

Gln Pro Lys Gln Glu Glu Gln Lys Thr Leu Asn Ile Ser Ala Thr Pro
100 105 110

Ala Pro Lys Gln Glu Gln Ser Gln Thr Thr Thr Glu Ser Thr Thr Pro
115 120 125

Lys Thr Lys Val Thr Thr Pro Pro Ser Thr Asn Thr Pro Gln Pro Met
130 135 140

Gln Ser Thr Lys Ser Asp Thr Pro Gln Ser Pro Thr Ile Lys Gln Ala
145 150 155 160

Gln Thr Asp Met Thr Pro Lys Tyr Glu Asp Leu Arg Ala Tyr Tyr Thr
165 170 175

Lys Pro Ser Phe Glu Phe Glu Lys Gln Phe Gly Phe Met Leu Lys Pro
180 185 190

Trp Thr Thr Val Arg Phe Met Asn Val Ile Pro Asn Arg Phe Ile Tyr
195 200 205

Lys Ile Ala Leu Val Gly Lys Asp Glu Lys Lys Tyr Lys Asp Gly Pro
210 215 220

Tyr Asp Asn Ile Asp Val Phe Ile Val Leu Glu Asp Asn Lys Tyr Gln
225 230 235 240

Leu Lys Lys Tyr Ser Val Gly Gly Ile Thr Lys Thr Asn Ser Lys Lys
245 250 255

Val Asn His Lys Val Glu Leu Ser Ile Thr Lys Lys Asp Asn Gln Gly
260 265 270

```
Met Ile Ser Arg Asp Val Ser Glu Tyr Met Ile Thr Lys Glu Glu Ile
        275                 280                 285

Ser Leu Lys Glu Leu Asp Phe Lys Leu Arg Lys Gln Leu Ile Glu Lys
    290                 295                 300

His Asn Leu Tyr Gly Asn Met Gly Ser Gly Thr Ile Val Ile Lys Met
305                 310                 315                 320

Lys Asn Gly Gly Lys Tyr Thr Phe Glu Leu His Lys Lys Leu Gln Glu
                325                 330                 335

His Arg Met Ala Asp Val Ile Asp Gly Thr Asn Ile Asp Asn Ile Glu
            340                 345                 350

Val Asn Ile Lys
        355
```

<210> 32
<211> 313
<212> PRT
<213> Staphylococcus aureus

<400> 32

```
Met Glu His Thr Thr Met Lys Ile Thr Thr Ile Ala Lys Thr Ser Leu
  1               5                  10                  15

Ala Leu Gly Leu Leu Thr Thr Gly Val Ile Thr Thr Thr Thr Gln Ala
            20                  25                  30

Ala Asn Ala Thr Thr Leu Ser Ser Thr Lys Val Glu Ala Pro Gln Ser
        35                  40                  45

Thr Pro Pro Ser Thr Lys Ile Glu Ala Pro Gln Ser Lys Pro Asn Ala
    50                  55                  60

Thr Thr Pro Pro Ser Thr Lys Val Glu Ala Pro Gln Gln Thr Ala Asn
65                  70                  75                  80

Ala Thr Thr Pro Pro Ser Thr Lys Val Thr Thr Pro Pro Ser Thr Asn
                85                  90                  95

Thr Pro Gln Pro Met Gln Ser Thr Lys Ser Asp Thr Pro Gln Ser Pro
            100                 105                 110

Thr Thr Lys Gln Val Pro Thr Glu Ile Asn Pro Lys Phe Lys Asp Leu
        115                 120                 125

Arg Ala Tyr Tyr Thr Lys Pro Ser Leu Glu Phe Lys Asn Glu Ile Gly
    130                 135                 140

Ile Ile Leu Lys Lys Trp Thr Thr Ile Arg Phe Met Asn Val Val Pro
145                 150                 155                 160

Asp Tyr Phe Ile Tyr Lys Ile Ala Leu Val Gly Lys Asp Asp Lys Lys
                165                 170                 175

Tyr Gly Glu Gly Val His Arg Asn Val Asp Val Phe Val Val Leu Glu
            180                 185                 190

Glu Asn Asn Tyr Asn Leu Glu Lys Tyr Ser Val Gly Gly Ile Thr Lys
        195                 200                 205
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```
Ser Asn Ser Lys Lys Val Asp His Lys Ala Gly Val Arg Ile Thr Lys
    210                 215                 220

Glu Asp Asn Lys Gly Thr Ile Ser His Asp Val Ser Glu Phe Lys Ile
225                 230                 235                 240

Thr Lys Glu Gln Ile Ser Leu Lys Glu Leu Asp Phe Lys Leu Arg Lys
                245                 250                 255

Gln Leu Ile Glu Lys Asn Asn Leu Tyr Gly Asn Val Gly Ser Gly Lys
            260                 265                 270

Ile Val Ile Lys Met Lys Asn Gly Gly Lys Tyr Thr Phe Glu Leu His
        275                 280                 285

Lys Lys Leu Gln Glu Asn Arg Met Ala Asp Val Ile Asp Gly Thr Asn
    290                 295                 300

Ile Asp Asn Ile Glu Val Asn Ile Lys
305                 310
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INTERNATIONAL SEARCH REPORT

International Application No
PCT/GB 01/02685

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C12N15/31 C12N15/63 G01N33/68 C07K14/31 A61K39/085
C07K16/12 C12N5/12 A61K39/40

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 C12N G01N C07K A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)
EPO-Internal, EMBL, WPI Data, BIOSIS

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	ARIFUR RAHMAN ET AL.: "Gamma-Hemolysin genes in the same family with LukF and lukS genes in methicillin resistant Staphylococcus aureus" BIOSCIENCE BIOTECHNOLOGY BIOCHEMISTRY., vol. 57, no. 7, 1993, pages 1234-1236, XP002177747 TOKYO JP the whole document ---	1-9, 18-48
A	WO 99 50418 A (NEUTEC PHARMA PLC) 7 October 1999 (1999-10-07) the whole document -----	1-9, 18-49

☐ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
18 September 2001	19. 11. 2001

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	MONTERO LOPEZ B.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/GB 01/02685

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

Although claims 26-32 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

2. [ ] Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1. [ ] As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. [ ] As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. [X] No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Partially 1-9, 18-49

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

International Application No. PCT/GB 01/02685

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. Claims: Partially 1-9, 18-49

Staphylococcus aureus antigen encoded by a DNA sequence of SEQ ID NO:1, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

2. Claims: Partially 1-9, 18-49

Staphylococcus aureus antigen encoded by a DNA sequence of SEQ ID NO:2, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

3. Claims: Partially 1-9, 19-46

Staphylococcus aureus antigen encoded by a DNA sequence of SEQ ID NO:3, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

4. Claims: Partially 1-9, 19-46

Staphylococcus aureus antigen encoded by a DNA sequence of SEQ ID NO:4, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

5. Claims: Partially 1-9, 19-46

Staphylococcus aureus antigen encoded by a DNA sequence of SEQ ID NO:5, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

6. Claims: Partially 1-9, 18-48

Staphylococcus aureus antigen encoded by a DNA sequence of SEQ ID NO:6, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

7. Claims: Partially 1-9, 19-46

Staphylococcus aureus antigen encoded by a DNA sequence of SEQ ID NO:7, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

8. Claims: Partially 1-9, 19-46

Staphylococcus aureus antigen encoded by a DNA sequence of SEQ ID NO:8, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

9. Claims: Partially 1-9, 19-46

Staphylococcus aureus antigen encoded by a DNA sequence of

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

SEQ ID NO:9, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

10. Claims: Partially 1-9, 18-48

Staphylococcus aureus antigen encoded by a DNA sequence of SEQ ID NO:10, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

11. Claims: Partially 1-9, 18-48

Staphylococcus aureus antigen encoded by a DNA sequence of SEQ ID NO:11, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

12. Claims: Partially 1-9, 19-46

Staphylococcus aureus antigen encoded by a DNA sequence of SEQ ID NO:12, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

13. Claims: Partially 1-9, 19-46

Staphylococcus aureus antigen encoded by a DNA sequence of SEQ ID NO:13, DNA sequence and variants thereof; vectors and host cells comprising the same and their use for the production of the polypeptide; vaccine comprising the

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

antigen and its use in immunisation; antibodies directed to the antigen and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament; use of the polypeptides for preparing a hybridoma cell-line.

14. Claims: 10-17, and partially 24-46

Method to identify antigenic polypeptides by transfecting a pathogenic organism gene library into a host cell and contacting the expressed polypeptides with autologous antisera from an animal infected with the pathogenic organism; polypeptides so obtained, vaccines comprising the antigenic polypeptides and use in immunisation; antibodies directed to the antigenic polypeptides and vectors and hybridoma cells for their production; use of the antibodies for the manufacture of a medicament.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/GB 01/02685

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9950418 A	07-10-1999	AU 3156699 A	18-10-1999
		EP 1068328 A1	17-01-2001
		WO 9950418 A1	07-10-1999